

Book of Abstracts

of the 75th Annual Meeting
of the European Federation of Animal Science



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**Book of Abstracts of the 75th Annual Meeting of the
European Federation of Animal Science**



EAAP

European Federation of Animal Science

The European Federation of Animal Science wishes to express its appreciation to the Ministero dell'agricoltura, della sovranità alimentare e delle foreste (Italy) and the Associazione Italiana Allevatori (Italy) for their valuable support of its activities.

Book of Abstracts of the 75th Annual Meeting of the European Federation of Animal Science

Florence, Italy, 1st – 5th September, 2024



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Welcome to the EAAP 2024 in Florence

Ladies and Gentleman,

on behalf of the organizing committee and of the whole community of the Italian Association for Animal Science and Production I am very happy and honored to welcome you to the 75th annual meeting of the European Association for Animal Production, that will take place in Firenze (Florence) from September 1st to September 5th 2024.

Firenze is one of the main art cities of Italy. An outstanding richness of monuments, historical sites, museums and churches represents the cultural and artistic heritage of Firenze, considered the capital of the Renaissance. Firenze is the capital of the region of Tuscany, placed in the heart of Italy, and known worldwide for its beautiful hilly agricultural Landscapes, the country mansions of the region of Chianti, the wines, the olive oil, the autochthonous giant beef cattle "Chianina", the "Fiorentina" steak. These products represent excellences of the Italian agriculture. Social events during the EAAP congress, hosted on some of the most spectacular venues of the City, will offer the opportunity to directly taste several of these products.

The general theme of the congress is "Global quality: Environment, Animals, Food". The congress will consist in several parallel scientific sessions where updated research results covering the whole domain of the Animal Science will be presented and discussed together with the presentation of main ongoing research projects. With its geographical position, Italy exhibits a great variety of environments, ranging from Alpine to Mediterranean climate. This peculiarity is reflected in the structure of its agriculture. Intensive dairy and beef cattle, pigs and poultry farms are typical of the Northern district. Buffalo farming can be found in Latium and Campania, in the Central-Southern part of Italy. Small ruminant systems characterize Southern Italy and the two main islands: Sicily and Sardinia. This richness results in a number of biodiverse livestock farming systems, which generate high-quality products, in many cases certified as PDO or PGI. The products are expression of local genetic resources and of the Italian cultural heritage and they contribute to define the "Made in Italy" brand. Some of these production systems will be the target of technical tours organized at the end of technical sessions.

Benvenuti a Firenze!

Presidents

Nicolò Pietro Paolo Macciotta

Marcello Mele

Industry Members Club



EAAP started in 2023 a new initiative to create closer connections between European livestock industries and the animal science network. Therefore, the “EAAP Industry Club” was shaped with the specific aim of bringing together the important industries of the livestock sector with our European Federation of Animal Sciences. All companies dealing with animal production (nutrition, genetic, applied technologies, etc.) are invited to join the “EAAP Industry Club” because industries will have opportunity to increase their visibility, to be actively involved in European animal science activities, and to receive news and services necessary to industries. In addition, through the Club, industries will enlarge their scientific network and will receive specific discounts on sponsoring activities.

The Industries that already joined the “EAAP Industry Club” are:



The Club gives:

Visibility • Company name and logo at EAAP website and all relevant documents • Slides with name and logo at Official Events • Priority links with EAAP Socials • Invite, through EAAP dissemination tools and socials, people to events organized by your company • Information disseminated through a brand new Industry Newsletter • Networking • Joining the Study Commissions and Working Groups • Suggest topics to be considered for Annual Meetings Scientific Sessions • Organize Professional Panel through the EAAP platforms • Economic Benefits • One free registration to each Annual Meeting and at every meeting organized by EAAP • Five individual memberships at no cost • Many possible discounts (-30%) to increase company visibility through: EAAP Newsletter, EAAP website, EAAP Annual Meetings and workshops • Support young scientist by sponsoring scholarships named by the company • Co-Organize and sponsor webinars

Make yourself more visible within the livestock industry via the animal science network!

For more information please contact eaap@eaap.org

YoungEAAP



What is the YoungEAAP?

YoungEAAP is a group of young scientists organized under the EAAP umbrella. It aims to create a platform where scientists during their early career get the opportunity to meet and share their experiences, expectations and aspirations. This is done through activities at the Annual EAAP Meetings and social media. The large constituency and diversity of the EAAP member countries, commissions and delegates create a very important platform to stay up-to-date, close the gap between our training and the future employer expectations, while fine-tuning our skills and providing young scientists applied and industry-relevant research ideas.

Committee Members at a glance

- Ines Adriaens (President)
- Jana Obsteter (Vice President)
- Giulia Gislon (Secretary)

YoungEAAP promotes Young and Early Career Scientists to:

- Stay up-to-date (i.e. EAAP activities, social media);
- Close the gap between our training and future employer expectations;
- Fine-tune our skills through EAAP meetings, expand the special young scientists' sessions, and/ or start online webinars/training with industry and academic leaders;
- Meet to network and share our graduate school or early employment experiences;
- Develop research ideas, projects and proposals.

Who can be a Member of YoungEAAP?

All individual members of EAAP can join the YoungEAAP if they meet one of the following criteria: Researchers under 38 years of age OR within 10 years after PhD-graduation.

Just request your membership form (ines.adriaens@kuleuven.be) and become member of this network!!!

EAAP Study commissions



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Sponsored talks

Sponsored by: Silvateam

September 1st at 15:45, Room: Maremmana

Silvafeed ATX to mitigate the effect of oxidative stress in animals' health and meat quality

Elizabeth Santin, DMV, PhD – esantin@silvateam.eu

Polyphenols are a diverse group of naturally occurring compounds found in plants, known for their potent antioxidant properties. In animal studies, polyphenols have demonstrated significant potential in mitigating oxidative stress, a condition characterized by an imbalance between the production of reactive oxygen species (ROS) and the antioxidant defense system. Oxidative stress is implicated in various pathological conditions, including inflammation, aging and chronic diseases. In livestock, oxidative stress can adversely affect meat quality, leading to lipid peroxidation, protein oxidation, and discoloration, which in turn reduce shelf life and consumer acceptability. Polyphenols exert their beneficial effects by scavenging free radicals, chelating metal ions and modulating antioxidant enzyme activities. Additionally, they influence cellular signaling pathways and gene expression related to oxidative stress responses. However not all polyphenols have the same anti-oxidant effect. We will present evidence regarding Silvafeed ATX, a well-balanced blend of polyphenols that stands out for its efficacy in improving antioxidant status, reducing markers of oxidative damage and enhancing meat quality. The mechanisms by which this specific blend of polyphenols counteract oxidative stress will be presented to discuss their potential applications in animal health management, disease prevention and improvement of meat quality.

Sponsored by: Tecnosens

September 2nd at 15:45, Room: Auditorium

Tecnosens Sniffer for CO2 and CH4 monitoring: Our experience in smart agriculture

Greta Bonacina, Tecnosens Sales Department - gbonacina@tecnosens.it

Established in 1994, Tecnosens has always been involved in the latest technologies for gas monitoring, sensing technology and equipment. Also, environmental issues have always been a Leitmotiv in Tecnosens R&D. As a contribution to livestock research, thanks to the cooperation with several research institutes and the environmental sensitivity, Tecnosens could develop MooLogger, a robust and reliable Sniffer to be installed in farms to monitor the breath of the ruminants composed mainly by CO2 and CH4. Tecnosens provides a wide range of sensors and solutions for farm emissions monitoring and environmental control, to monitor NH3 and other pollutants, for several farming and agriculture application: discover which solution may suit your needs.

Sponsored by: MSD

September 3rd at 10:15, Room: Sarda

Technology for farms: a crucial turning point for the sector

Massimiliano Intini - massimiliano.intini@msd.com

The introduction of technology in farms is a topic of crucial importance for the agricultural sector and represents a turning point for the sector for dairy cattle farming. We, at MSD Animal Health, feel responsible for promoting innovations that guarantee the sustainability of production and safety for consumers. Advanced monitoring systems allow real-time control of the health of cows, optimizing the use of resources and improving animal welfare. Sophisticated sensors can accurately detect vital parameters, allowing timely and targeted interventions. This not only increases productivity but also reduces the risk of diseases, improving the quality of the milk produced. Sustainability is another fundamental pillar. Thanks to innovative technologies, we can minimize the environmental impact of our activities. Advanced management systems allow more efficient use of water and energy, reducing waste and cutting greenhouse gas emissions. In this way, we not only respect the environment but also meet the needs of an increasingly attentive and aware consumer. Speaking of consumers, it is essential to ensure them maximum transparency and safety. Traceability technologies allow us to follow every stage of the production process, from field to table, respecting the highest quality standards. Consumer trust is our most precious resource and technology is the tool that allows us to earn it every day. In conclusion, the introduction of technology in Italian dairy farms is not just an option, but a necessity: it is a path that we must follow with determination and vision, aware that every step forward is an investment in our future.

Sponsored by: Vetagro

September 3rd at 12:45, Room: Sarda

From mitigating environmental impact to antimicrobial reduction: how botanicals can improve animal production sustainability

Benedetta Tugnoli - benedetta.tugnoli@vetagro.com, Richard Paratte - richard.paratte@vetagro.com

The goal of our talk is to explore the potential of botanical compounds and organic acids to improve the sustainability of animal production. Two experts, one specialized in ruminant species and the other in monogastric species, will guide you through this analysis.

Key challenges

We will focus on two key challenges:

- Environmental impact: the use of functional nutritional strategies allows us to improve the metabolism of farmed animals in order to reduce its environmental impact.
- Antimicrobial resistance: Excessive use of antibiotics in animals threatens human and animal health.

Potential of botanical compounds

Botanical extracts offer various beneficial properties:

- Antimicrobial: fight pathogens and reduce the need for antibiotics.
- Antioxidants: protect cells from oxidative damage.
- Immunomodulatory: strengthen the immune system of animals.

Applications

The use of microencapsulated botanical compounds in animal diets has been shown to promote gut health, improve nutrient utilization and reduce dependence on antibiotics.

Our experts will delve into the specific applications of botanical compounds for:

- Ruminants: dairy cows
- Monogastric: pigs, poultry

Conclusion

Botanical compounds can play a key role in making animal production more sustainable. Using these solutions can reduce environmental impact, improve animal health and help fight antimicrobial resistance.

Question and answer session

At the end of the presentation, there will be the opportunity to ask questions to our experts.

Sponsored by: Illumina

September 4th at 15:45, Room: Auditorium

Genomics in Agriculture: Advancing Productivity and Sustainability through a One Health Framework

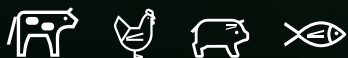
André Eggen: aeggen@illumina.com

The integration of genomics into the agricultural and food industries has ushered in a new era of innovation, promising to enhance productivity, sustainability, and food safety from farm to fork. Adopting a One Health framework ensures that the use of genomics tools considers the broader ecological impacts, promoting sustainable practices. For instance, reducing the prevalence of antimicrobial resistance (AMR) benefits both human and animal health. Furthermore, monitoring environmental DNA (eDNA) can assess the impact of farming practices on biodiversity, guiding more eco-friendly approaches.

Research is written in our DNA



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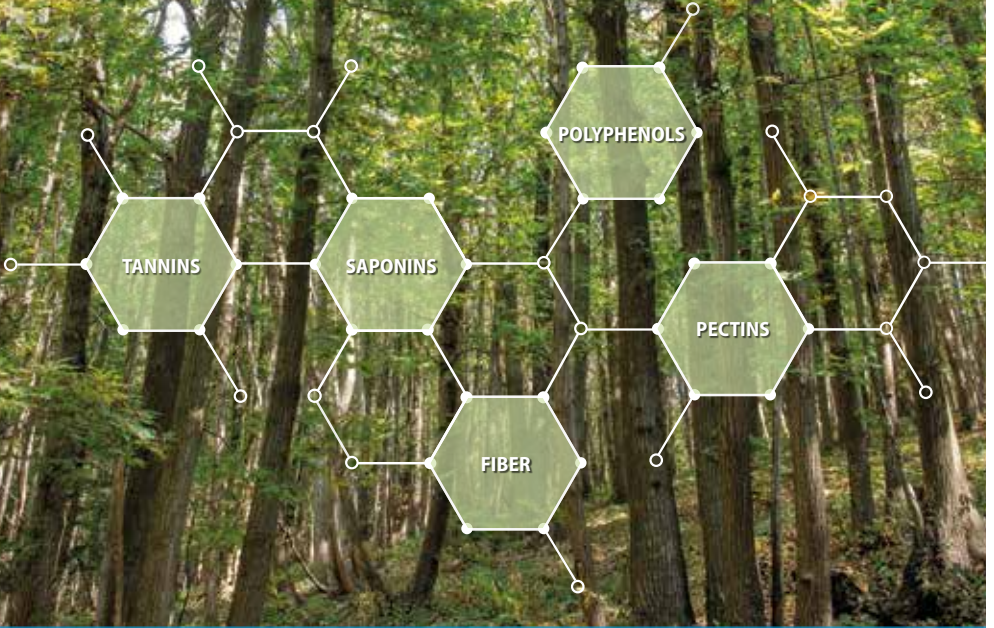


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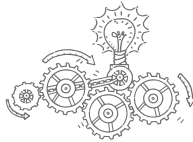
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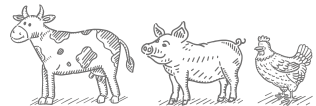
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Our mission is to re-introduce several **natural compounds** lost due to modern breeding systems, showing how to maximise their benefits during the daily life of any livestock species. These eco-designed additives and premixes can provide **holistic solutions** to target critical issues related to environmental footprint, nutrition, physiology and balance between performance and wellbeing.



Ruminants breath and environmental gas *monitoring* are essential to develop a *sustainable future* for our planet:

Tecnosens works with Research institutes and Companies providing *gas sensing solutions* for Smart Agriculture field



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- ▣ Reduction of ingested dry matter
- ▣ Improvement of animal welfare



VetosEurope was founded by combining the expertise and savoir faire of one of the most important families in the world in live cattle trade and the technical and scientific knowledge of a group of scientists and researchers specialized in the research and development of **natural feed supplements and nature-identical substances for cattle**, aimed at increasing production while at the same time reducing CH_4 and CO_2 emissions.

The research into an innovative technology aimed at protecting the environment by using nature has led VetosEurope to create **ANAVRIN®**.

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VetosEurope



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Per questo la ricerca scientifica di MSD Animal Health è impegnata a offrire trattamenti innovativi e a promuovere azioni concrete che proteggano gli animali, le persone e il nostro pianeta attraverso prevenzione, sostenibilità, sicurezza alimentare, tracciabilità, cura e benessere.

Scientific programme

EXPLANATORY NOTES ON SESSIONS

Abbreviations after the title are (Organising Commission; Session Type)

Commissions are:

Cattle (C), Genetics (G), Health and Welfare (H+W), Horse (H), Insect (I), Livestock Farming Systems (LFS), Nutrition (N), Pig (P), Physiology (Ph), Precision Livestock Farming (PLF), Sheep and Goat (S+G), Animal Task Force (ATF)

Session types are:

Theme Sessions (T) are on key topics in animal science and consist of invited and offered papers.

Industry sessions (I) are led and supported by Industry and comprise invited and offered papers.

Free communications/Bottom-Up (B): these sessions will be created from submitted abstracts and titles will be announced in early April.

Discovery Sessions (D) are invited single/multiple presentations on hot/emerging topics.

Challenge Sessions (C) are free format (workshop, open committee, round table) to debate important issues, not theatre paper presentation.

Early-career (EC) scientist's Session.

Project session (P) are sessions based around a project which wants to share its results, often combined with submitted abstracts.

Working Group sessions (WG) are the results of an EAAP working group.



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Room	Sunday 1st Sept 8.30 – 12.30	Sunday 1st Sept 14.00 – 18.00
Auditorium Palazzo Congressi Lower Ground Floor	1. Integrating mitigation and adaptation breeding strategies (G, T)	15. Role of bioinformatics applied to livestock data - exploiting structural variation and pangenome-based techniques for livestock (G, T)
Pisana Palazzo Congressi 2nd Floor	2. Breeding scheme optimization: balancing breeding goal(s), genetic progress and diversity (G, T)	16. Genomic selection tools for within- and across- herd management in livestock species (G, T)
Valdostana Palazzo Affari 3rd Floor	3. N, P and C efficient use and circularity in dairy farms – high utilization, minimal losses (C, T)	17. Global quality of animal products in 2024: stakeholders' perceptions, desires, and priorities in a changing world (C, P)
Reggiana Palazzo Affari 2nd Floor	4. Research on young stock care: the journey of calves from gestation onwards (C/H+W, T)	18. Sustainable and smart integration of the dairy and beef sectors in EU (C/CODABE, T)
Chianina Palazzo Affari 1st Floor	5. Innovative and emerging feed and forage resources (N, T)	19. Nutritional models development and applications in livestock farming (N, T)
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Romagnola Palazzo Affari 1st Floor	7. Emerging practices and tools in horse production (H, BU)	21. Heat stress in pig and poultry production: consequences and strategies to cope with global warming (P, T)
Piemontese Palazzo Affari 2nd Floor	8. Innovative approaches to pig and poultry production (P, EC)	22. Innovation in pig genetics (P/G, B)
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Rendena Palazzo Affari 4th Floor	11. Insect nutrition (I, T)	25. Project session ADVAGROMED (I, P)
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Modicana Palazzo Affari 3rd Floor	13. Sensing physiology: Tools towards optimising livestock husbandry (Ph, T)	20. Functional feed additives in poultry nutrition (N/PWG, WG)
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Valdostana Palazzo Affari 3rd Floor		32. The role of marbling in beef quality – development, importance, measurement, harmonisation (C, T)
Reggiana Palazzo Affari 2nd Floor		33. Mediterranean buffalo farming for sustainable milk and meat production (C, T)
Chianina Palazzo Affari 1st Floor		34. Nutrition management to reduce methane emissions and environmental impact, Part 1 (N, T)
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Romagnola Palazzo Affari 1st Floor		36. Slow-growing meat-type and dual purpose genotypes for conventional and alternative farming systems in the EU (PWG/WPSA-Italian Branch, WG)
Piemontese Palazzo Affari 2nd Floor		37. Rethinking the transition from suckling to weaning to prevent negative consequences of the stress on pig health (P, T)
Podolica Limonaia - Garden		38. Best-practice in addressing pig welfare on-farm (H+W, I)
Marchigiana Palazzo Affari Ground floor		39. Building quality into animal products to improve the sustainability of farming systems for the future (LFS, T)
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Calvana Palazzo Congressi Ground floor		40. Zooarchaeological research lessons for contemporary livestock management, conservation and genetics (S+G/LFS, T)
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Valdostana Palazzo Affari 3rd Floor	45. Indicators, hi-tech solutions, and tools to effectively manage cattle (C, T)	59. Milk properties for the valorization of dairy (by-products) manufacturing (C/ADSA, T)
Reggiana Palazzo Affari 2nd Floor	46. Viable future dairy farming systems and products from production, feeding, environmental and consumer viewpoint, Part 1 (C, P)	60. Viable future dairy farming systems and products from production, feeding, environmental and consumer viewpoint Part 2 (C, P)
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Romagnola Palazzo Affari 1st Floor	49. Nutrition in health and welfare of ruminants (N/H+W, T)	63. Improving animal welfare by optimising nutrition/feeding behaviour, Part 1 (H+W/N, T)
Piemontese Palazzo Affari 2nd Floor	50. Sustainable solutions to support and sustain gut health in monogastric livestock (with project MONOGUTHEALTH), Part 1 (P, P)	64. Sustainable solutions to support and sustain gut health in monogastric livestock (with project MONOGUTHEALTH), Part 2 (P, P)
Podolica Limonaia - Garden	51. Genetic progress vs animal welfare? (H+W/G/FABRE-TP, C)	65. PLF for health, behaviour and welfare, Part 2 (H+W/PLF, T)
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Reggiana Palazzo Affari 2nd Floor	74. From horse welfare to social license to operate (H, T) <i>Horse Commission Business Meeting</i>	88. How the dairy sector copes with EU-Green deal (C, T)
Chianina Palazzo Affari 1st Floor	75. Advances in non-ruminant nutrition, Part 2 (N, T) <i>Nutrition Commission Business Meeting</i>	89. Free communications in animal nutrition (N, BU)
Maremmana Palazzo Affari 2nd Floor	76. Advances in ruminant nutrition, Part 2 (N, T)	90. Nutrition and feeding in the circular economy (N, T)
Romagnola Palazzo Affari 1st Floor	77. Improving animal welfare by optimising nutrition/feeding behaviour, Part 2 (H+W/N, T)	91. Young EAAP session. Project writing: tools, opportunities and experiences (All, EC)
Piemontese Palazzo Affari 2nd Floor	78. Collaboration in experimental research for sustainable pig production (with PIGWEB) (P, P) <i>Pig Commission Business Meeting</i>	92. Good practices in experimental research for sustainable pig production (with PIGWEB) (P, P)
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Marchigiana Palazzo Affari Ground floor	80. Technologies for GHG emission mitigation on farm: options, opportunities and challenges (LFS, T) <i>LFS Commission Business Meeting</i>	94. Living labs and demonstration farms: approaches to improve sustainability of LFS globally (LFS, T)
Rendena Palazzo Affari 4th Floor	81. Safety, sustainability and welfare in the insect (I, T) <i>Insect Commission Business Meeting</i>	95. Insects as food & feed (I/PWG, T)
Calvana Palazzo Congressi Ground floor	82. Optimization of pasture forage and by-product resources to improve sheep and goat production (S+G, T) <i>Sheep + Goat Commission Business Meeting</i>	96. Optimization of technical, economic, and environmental efficiency in sheep and goat production systems (S+G, T)
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Scientific programme

Session 1. Integrating mitigation and adaptation breeding strategies

Date: Sunday 1 September 2024; 8:30 - 12:30

Chair: Sell-Kubiak / Gredler-Grandl

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Chair: Berg / Rind Thomassen

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Chair: Kuipers / Ruska

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Chair: Costa / Vestergaard

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Chair: Bellezza Oddon / De Smet

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Chair: Hadjipavlou / Ligda

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Chair: Atzori / Munoz

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Chair: Kreuzer-Redmer / Camarinha Silva

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Chair: Chalvon-Demersay / Stadnicka

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Chair: Verschuren / Nilsson

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Chair: Lewis / Lopreiato

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Chair: Lee / O'Mara

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Chair: Rumbos / Biasato

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Chair: Nagy

Theatre Session 26

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Chair: Schiavo / Pocrnic

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Chair: Aernouts

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Chair: Galama / Brocard

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Chair: Bruggeman / Pinotti

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Date: Tuesday 3 September 2024; 8:30 - 12:30

Chair: Tan / Mantovani

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Chair: Kreuzer-Redmer / Foris

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Date: Tuesday 3 September 2024; 8:30 - 12:30

Chair: Bee / Trevisi

Theatre Session 50

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Chair: Gottardo / Duijvesteijn

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Chair: Pouloupoulou / Sturaro

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Chair: Sandrock / Petersen

Theatre Session 53

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Chair: Kiefer / Keogh

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Date: Tuesday 3 September 2024; 14:30 - 17:00

Chair: Maselyne / Odintsov Vaintrub

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Chair: Egger-Danner / Tenhunen

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Chair: Frigga / Probo

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Chair: Gottardo / Kreuzer-Redmer

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Chair: Bee / Millet

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Chair: Almeida / Campion

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Chair: Salami

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Chair: Deruytter / Smetana

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Chair: Tsiplakou / Rahmatalla

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Chair: De Grande / Druyan

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Chair: Kuipers / Maggiolino

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Chair: Klopčič / Cieslak

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Chair: Ottoboni / Kar

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Date: Wednesday 4 September 2024; 14:00 - 14:15

Chair: Adriaens / Obsteter / Gislou

Theatre Session 91

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Date: Wednesday 4 September 2024; 14:00 - 17:45

Chair: Bee / Millet

Theatre Session 92

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Chair: Minozzi / Holinger

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Session 94. Living labs and demonstration farms: approaches to improve sustainability of LFS globally

Date: Wednesday 4 September 2024; 14:00 - 18:00

Chair: Sturaro / Rivero

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Chair: Boatta / Naser El Deen

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Session 1

Theatre 1

Global Methane Genetics: a global program to accelerate genetic progress for reduced methane emission in ruminants

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Methane (CH₄) emissions have been identified as major contributor to global warming. In livestock, various CH₄ mitigation strategies are heavily researched, such as management and feeding strategies or genetic selection. Genetic selection is a reliable, cost-effective, cumulative and permanent mitigation tool. Despite several countries having started to record individual CH₄ emission of cattle and small ruminants, it is still far from routine implementation, given the currently small number of phenotypes. The Global Methane Genetics (GMG) program aims to accelerate genetic progress towards low-CH₄ emitting ruminants in the Global North and South by (1) enhancing harmonisation of recording and trait definition by shared protocols, SOP and existing data in the public domain (2) expanding methane recording and phenotype collection globally (3) developing efficient breeding programs for CH₄ reduction, and (4) implementation of routine genetic evaluations. The GMG program and global activities are centred around seven working groups, database development, new research and recording of methane emission in cattle and small ruminants and involves key livestock industry segments. The working groups are assigned to identify gaps and needs and develop research proposals for global dairy and beef cattle, small ruminants and buffalos. Specific working groups for Africa, Asia and South America address regional peculiarities in recording of CH₄ emission. The program conducts impact analysis across all working groups to show the potential of animal breeding as CH₄ emission mitigation tool.

Session 1

Theatre 2

Are the most productive, the less thermotolerant animals in low input beef cattle populations? The case of the Avileña-Negra Ibérica beef cattle breed

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Climate change is imposing many distresses in livestock populations. Many studies have been performed on dairy species where trade-offs are described between production level and the susceptibility to heat stress. On the contrary, little attention has been paid to low input beef cattle populations under extensive systems. Our objective was to characterize the effect of heat stress on weaning weight (WWT) of calves and age at first calving (AFC) of cows of ANI breed, as well as to estimate trade-offs. WWT and AFC records of 55,248 and 22,779 calves and cows were used for the analysis, respectively. Average (Tave), maximum and minimum daily temperatures (°C) collected from weather stations were used as measures of heat load (HL). For WWT, lags were HL at day of weighing and average HL up to 30 d prior to weaning. For AFC, HLs were studied around conception (± 30 days), assuming a gestation length of 285 days. Reaction norm models under the so called “broken-line” model were applied. Thus, we first estimated the temperature change points at which the animals began to show some response to thermal stress. The best models were those including Tave for a period of 30 d (Tave30) for WWT and 7 d post-conception (Tave7post) for AFC. Change points were 22°C for WWT and 25°C for AFC. Slopes were negative for WWT. and positive for AFC but, as modeled, the impact of heat stress on AFC was negligible. Genetic correlation between the intercept and slope of change in WWT with HL was high and positive, indicating that animals that weigh more are those coping better with heat stress. This work is funded by the EU’s Horizon Europe research and innovation programme under the grant agreement No 01059609 (Re-Livestock)

Feed efficiency, performance, and methane emissions of finishing Hereford steers with high and low residual feed intake

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Feed efficiency effect was measured by residual feed intake (RFI), on animal performance, enteric methane emission (CH₄), yield (CH₄-Y), and intensity (CH₄-I). RFI groups were defined by half standard deviation from the average, above, the efficient (LRFI), and below, the inefficient (HRFI) steers. Data from 136 Hereford steers was recorded from two RFI tests (T1, T2) during fattening, with a total mixed ratio that included a fiber source, corn grain, and a mineral supplement. In T1 67 steers were fed with sorghum silage as a fiber source, while in T2 maize silage was used with 69 animals. Feed intake was recorded with automatic feeders (Growsafe-Vytelle) in two 70d feed efficiency tests, and CH₄ was measured with GreenFeed system (C-Lock). Recruitment for CH₄ were 64% (T1, n=45) and 83% (T2, n=58). A linear regression model was used for estimating RFI, including dry matter intake (DMI), average daily gain (ADG), metabolic weight (MW), and fat thickness (FT) by ultrasound at the end of the test. The comparison of LRFI and HRFI showed non-significant differences for ADG, MW, and FT ($p > 0.05$), but LRFI steers ate less feed (10.6 vs 12.6 kg DM, $p < 0.05$). As expected, DMI and CH₄ were associated with correlations that vary between diets ($r = 0.43$ in T1 and $r = 0.33$ in T2). Efficient animals (LRFI) ate 19% lower DMI per day, with only a 6% reduction of CH₄ which was associated with higher CH₄-Y (20.5 vs 18.6 g CH₄/kgDM, $p < 0.05$), compared to HRFI animals. The CH₄-I was also lower in LRFI animals (147 vs 157 gCH₄/kg, $p < 0.05$). Improving RFI contributes to economic benefits by reducing DMI without compromising production, and with a favorable effect on CH₄ and CH₄-I.

Session 1

Theatre 4

Trait definitions for methane production in Australian beef cattle

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Selection for lower methane (CH₄) emission in ruminants can be an efficient strategy to mitigate greenhouse gas emission from agriculture. Large-scale recording of CH₄ production (g/day) from beef cattle is currently occurring in Australia using GreenFeed Emission Monitors to obtain repeated measurements on grazing heifers and feedlot steers. The aim of this study was to determine the optimum trait definition for mean CH₄ production in terms of number of records per animal and minimum recording duration. After removing outliers (individual mean ± 3 SD), 82,226 records from 556 grazing heifers and 751 feedlot steers were available. The phenotypes were adjusted for diurnal pattern by subtracting the fixed effect of location-month-hour. Mean CH₄ production traits were then calculated using the within animal mean of animals with at least 1, 5, 10, 15, 20, 25 or 30 repeated measurements of 2- or 3-minute recording duration. The 14 traits were analyzed within diet by fitting mean CH₄ = population mean + breed + contemporary group + animal effect ($N(0, \sigma_a^2 \square G)$) + regressions on age (days) and trial weight (kg) + residual ($N(0, \sigma_e^2)$) using DMU. Estimated heritabilities (h^2) were 0.17-0.44 (SE 0.14-0.22) for heifers on pasture and 0.40-0.55 (0.15-0.21) for feedlot steers. The h^2 was higher in the feedlot steers when using records of at least 3-minute duration. In the grass-fed heifers h^2 was highest when animals had 10+ records of at least 3-minute duration. The recommendation is therefore to use records of animals with at least 1 record of 3-minute duration for animals recorded in a feedlot environment and at least 10 records of 3-minute duration for animals in grazing environments when calculating mean CH₄ production.

Genotype by environment interaction for growth traits in the Italian Limousine population

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Precisely measuring heat stress and elucidating the genetic basis of heat tolerance in animals represent key challenges in modern animal breeding. This is because environmental factors play a substantial role in influencing the growth and overall health of animals. This study aimed to investigate the genetic basis of tolerance to heterogeneous environmental conditions and the impact of heat stress on genetic parameters in the Italian Limousine beef cattle. Three growth traits were evaluated: average daily gain (ADG), weaning weight (WW), and yearling weight (YW). Environmental covariates were combined with data on animals raised in Italy between 1991 and 2022. After data editing, 108,205, 100,058, and 24,939 animals were used to analyze ADG, WW, and YW, respectively. The models included 4617, 4670, and 2048 genotyped animals for each trait counted. The presence of genotype by environment interaction (G×E) was evaluated through a multiple-trait model. Variance components were obtained, and differences were noted for the traits across diverse environments. ADG and YW additive genetic variances were lower at bad conditions, but higher estimates were obtained for WW in extreme environments. A small difference in heritability was observed among the environments for ADG, WW, and YW. Heritability for WW remained constant, whereas, for ADG and YW, reduced under harsh environmental conditions. These results suggest the possibility of obtaining different responses to selection for these traits depending on the environmental conditions. Overall, the results confirm the presence of G×E for growth traits in the studied population.

Session 1

Theatre 6

Future emission reductions deliverable by Irish beef and dairy breeding indices

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Agriculture represents the largest source of greenhouse gas emissions in Ireland, with roughly two-thirds of such emissions deriving from livestock production (i.e., enteric fermentation, manure management and artificial fertiliser application). Selection of genetically superior animals using breeding indices, alongside the cumulative and permanent nature of genetic selection and high adoption rates, is recognised by the Irish marginal abatement cost curve to deliver reduced emissions in a cost-effective manner. Results herein will present the long-term expected emissions reductions, as well as the associated industry-level economic benefits, derived from the Irish breeding indices used to select the most profitable beef and dairy replacements, and the most profitable beef animals for slaughter. A selection index theory approach, alongside variance components and index weightings provided by the Irish Cattle Breeding Federation, were used to determine index accuracies and variances, and the expected trait changes per unit change in each index. These outputs were then applied in a recursive geneflow model to estimate the industry-level economic benefits in the form of net present values of annualised benefits from genetic improvement over time. These results provide an estimate of the environmental benefits deliverable by the current breeding indices in their use to select genetically superior animals. Moreover, outputs from this study provides a point of comparison for the extent that alterations in the current breeding framework, such as increased selection intensity or the addition of new traits to indices, could deliver greater accumulation of environmental benefits.

Exploring different definitions of methane concentration phenotypes in dairy cattle

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Due to the 2030 climate targets, where the EU aims for a 55% reduction in greenhouse gas emissions, several countries are monitoring enteric methane (CH₄) from livestock. One of the mitigation options is to identify and selectively breed low-emission animals. A widely employed phenotyping method involves breath sampling, by a device commonly referred to as a “sniffer.” This device samples breath during milking, reporting CH₄ concentration in parts per million. While CH₄ concentration (MC) has been proposed as an indicator for CH₄ emissions (CH₄ g/d) due to its high correlation, there’s a lack of consensus on which phenotype should be used for estimating the breeding value. This study aims to 1) investigate 3 distinct phenotypes across three countries: a) average MC during lactation (MCavg), b) sum of the average of the 2 top values within each peak (MCsa2p), c) number of peaks (MCnop), 2) Estimate genetic parameters for these 3 phenotypes, including genetic correlations. The dataset consist as follow 132,961 records from 7,669 cows (Netherlands), 16,924 records from 3,426 cows (Spain), and 34,359 from 483 cows Poland. This work has received funding from the European Union’s Horizon Europe research and innovation programme under the grant agreement No 01059609 (Re-Livestock project) and BO-43.10-002 B1 Klimaat: Emissie reductie methaan veehouderij.

Session 1

Theatre 8

Relationships between methane emissions and milk production traits, an unfinished story

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Conventional wisdom holds that methane emissions tend to rise with increased milk yields in dairy cows which appears to be logical as higher milk production necessitates greater feed consumption. However, even if we can assume a positive relationship between milk and feed intake, there are trade-offs between milk yield and methane production, as well as the influence of feed efficiency on methane emissions that have to be taken into account. The main aim of this study was to investigate the linearity of relationships between methane emissions and milk production traits on a genetic level as currently published variance component estimations tend to produce not always very clear results. Knowledge of this is of high importance for residual (or efficient) methane trait definitions and multi-trait production-methane models. A large database of MIR-predicted methane emissions was available for Walloon Holstein cows (1,529,282 test-day records on 229,465 first-parity cows, 1,062,013 test-day records on 151,726 second-parity cows, 642,735 test-day records on 90,484 third-parity cows). The study was based on sire-daughter groups. Using random regression single-trait three-lactation test-day models GEBV for methane ($h^2 = 0.14$ for average methane across lactations) but also for milk, fat or protein content and yields were computed for sires with at least 30 daughters (969 bulls with a mean of 108 and median of 61 daughters for methane). Mean reliability for average methane for these bulls was 0.71 ranging from 0.52 to 0.98. After grouping GEBV in 3 equal-sized classes ranging from very low to very high reference trait GEBV (milk, fat, or protein content and yields) curvilinearity in the response of methane GEBV was clearly detected. When fitting a daughter number weighted GEBV regression from methane on milk, fat, and protein yields, an adjusted R^2 of only 0.065 was observed. When letting a Stepwise algorithm find the best combination of the five milk production traits (linear and quadratic) using the AIC criteria, all variables, except protein content remained in the model (adjusted $R^2 = 0.24$). The obtained results were clear indications of nonlinear genetic relationships, but the reasons for, or consequences of these are unknown.

Estimation of heritability and genetic correlation of methane phenotypes and yield traits in the Spanish Holstein population

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Methane from livestock accounts for 6.3% of anthropogenic greenhouse gas emissions. One strategy to mitigate its effects is the genetic selection of animals with low methane emissions. Sniffer device is becoming popular in this effort because it is an accurate non-invasive method, and allows large-scale recording of individual methane emission. In dairy farms, it is usually attached to the milking robots and collect the gases released during the eructation. In this study, different phenotypes of methane were compared, obtaining heritability values and genetic correlations among them and with yield traits (milk, protein and fat). Data obtained from 3426 dairy cows in 34 spanish commercial farms were analyzed to calculate meanCH₄, ratio of meanCH₄/CO₂, sum of peaks CH₄, mean of peaks CH₄, sum of max peaks and area under the curve. Further, meanCH₄ and sum of peaks CH₄ were also calculated using one measurement every 5 seconds for comparison. Methane traits showed h² ranging from 0.01 to 0.15. Genetic correlations among methane traits showed wide variability, but it was low to moderate with yield traits (-0.05 to 0.16). Definition of suitable methane phenotypes with moderate h² allows implementation of genetic selection to mitigate the methane emission in cattle. Acknowledgment: Horizon-Europe Re-Livestock project (101059609)

Genetic correlations between feed intake and GHG emissions measured on young bulls with production and reproduction traits measured in Italian Holstein cows

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Aim of this study was to investigate how productive and reproductive performance of Italian Holstein cows would be affected by selecting for improved feed efficiency and reduced greenhouse gasses emissions of young bulls. The study involved 221 candidate young bulls kept at the Genetic Centre of Italian National Breeders Association for the Holstein, Brown and Jersey dairy cattle breeds (ANAFIBJ) and phenotyped using the GreenFeed system and the Roughage Intake Control system. In addition, we included 1,686 cows from commercial farms and phenotyped for production and reproduction during routine data collection (e.g., milk yield, protein yield, days from first to last insemination, body condition score, predicted body weight). Cows were included in the analyses as full-sisters, paternal or maternal half-sisters of the candidate young bulls. All the individuals were genotyped using different SNP panels and then imputed to a medium density panel. Results showed moderate positive correlations between CH₄ and CO₂ emissions in bulls and milk, fat, protein, and lactose yield in full/half-sisters. Similar correlations were found for feed intake and feeding duration. All the correlations reached the values of 0.70 at most, suggesting the possibility to increase yield in the cows while controlling intake and carbon emissions in the young bulls. Similarly, correlations showed the possibility of improving cow fertility while also reducing feed intake and carbon emissions in the bulls.

Genetic parameters for methane production, intensity, and yield predicted from milk mid-infrared spectra throughout the lactation in Montbéliarde cows

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Genetic selection of dairy cows against methane (CH₄) emissions requires estimating genetic parameters and correlations with milk traits. Predicting CH₄ from mid-infrared (MIR) milk spectra provides sufficient data for genomic predictions. In this study, we used 608,072 MIR milk spectra collected from 92,500 first and second parity Montbéliarde cows from 70 to 200 days in milk to predict CH₄ emissions expressed as direct CH₄ production (MeP_direct) in g/d, CH₄ intensity (MeI) in g/kg of fat- and protein corrected milk (FPCM), and as CH₄ yield (MeY) in g/kg of dry matter intake. An indirect CH₄ production (MeP_indirect) in g/d was also obtained by multiplying MeI by the observed FPCM. Bivariate random regression models were performed between first and second parity data for a given CH₄ trait or between CH₄ traits and milk traits (milk yield [MY], fat [FY] and protein [PY] yields). Methane traits were stable both within and across lactations, with average genetic correlations greater than 0.93. Average heritability estimates ranged from 0.27 to 0.44. The four CH₄ traits were genetically distinct, as their average correlations were lower than 0.41 except for the correlations of 0.74 between MeP and MeY, and the correlation of 0.53 between MeI and MeP_indirect. All three MeP_direct, MeI and MeY presented weak correlations with MY (<0.27 in average), FY (from -0.16 to 0.04 in average), and PY (from -0.07 to 0.32 in average). On the contrary, MeP_indirect was highly correlated with the three milk traits, with average correlations greater than 0.73. We recommend selecting on a combination of CH₄ traits, to avoid any strong opposition between CH₄ and milk traits.

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Sequence-based GWAS of heat tolerance traits in Holstein and Montbeliarde cattle

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Global warming will affect European cattle production systems, decreasing overall performance and welfare. In such a context, it is thus relevant to identify genomic regions involved in heat tolerance for production and functional traits. By combining the French bovine genetic database (focusing on French Holstein and Montbéliarde dairy cattle) with weather conditions (Safran database, Meteo-France), we estimated breeding values (BV) of animals along a temperature-humidity index (THI) gradient with a reaction-norm model. Heat tolerance traits were defined as the slope of BV at THI 70 for three traits: test-day milk yield, conception rate at first insemination, and somatic cell score. 50K SNPs from 4564 Holstein and 1737 Montbéliarde bulls were imputed to whole genome sequence using the run9 of the 1000 bull genomes project, resulting in ~13 million SNPs (R_2 Minimac ≥ 0.2 and MAF ≥ 0.01) to perform within-breed GWAS of heat tolerance for the three traits. Few genomic regions were significantly associated with heat tolerance, however these regions differed between breeds and traits. Beyond revealing novel regions associated to heat tolerance, our study also identified some regions already reported in previous studies, emphasizing their relevance. Finally, given the genetic background of heat tolerance in both production and functional traits, selection for heat tolerance should be considered. This study received funding from the European Union's Horizon 2020 research and innovation program under grant number 101000226 (Rumigen) and from APIS-GENE (CAICalor). The authors thank Meteo-France for the Safran database.

Drafting the genomic landscape of North African sheep and chicken adaptation to harsh environments
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The PRIMA SCALA-MEDI project aims to characterize the genetic and phenotypic diversity of indigenous sheep and chicken breeds in the Mediterranean region and to investigate their adaptability to challenging environments. The project focuses on remote phenotyping for traits related to adaptation, genotyping, and exploring genome methylation patterns in animals raised under different environmental conditions. Data and samples have been collected from local breeds in Tunisia, Algeria, and Morocco, together with socio-economic information along sheep and chicken value chains. To date 861 sheep from 20 breeds and 767 local chickens have been genotyped with the multispecies IMAGE001v2 array that contains 8,258 SNP for chicken and 11,787 SNP for sheep. Genetic diversity, selection signatures and association to climate variables have been investigated to draft a genomic landscape of North-African sheep and chicken adaptation to harsh environments.

Session 1

Poster 14

Characterization of a new heat tolerance phenotype in dairy cattle based on automatic milking systems and mid-infrared spectra

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In recent years, breeding for heat tolerance in dairy cattle has become increasingly important to ensure good animal welfare and to reduce milk yield losses in stressful conditions such as heat waves. We here propose novel phenotypes for heat tolerance that utilize routinely generated data including automatic milking systems with daily measurements of milk yield and body weight, and monthly generated mid-infrared spectra. Within the Horizon Europe project Re-Livestock (GA No. 01059609), data from 1,478 farms including 677,318 cows from 2013 to 2021 across the Netherlands was used to estimate the effects of THI on various traits. For this, a kernel regression-based framework was developed to allow for a less rigid modelling than traditional reaction norm models including interaction effects to days-in-lactation, season, and parity. Individual heat tolerance was subsequently characterized as the relative impact of heat stress on a specific animal compared to estimated population-wide parameters, with the resulting heat resilience traits for F% and P% having an estimated heritability of 0.25 & 0.16. This subsequently results in an animal that is showing average performance at THI 50, yielding 0.36% above average F% and 0.19% above average P% at THI 70 in case it is one genetic standard deviation about population average for the respective heat resilience trait.

Genetic parameters of feed efficiency, predicted methane-related, and carcass traits during fattening period in Japanese Black cattle

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The objective of this study was to estimate heritabilities for feed efficiency, predicted methane-related, and carcass traits and the genetic correlations among these traits during fattening period in Japanese Black cattle. We used a total of 649 animals, which were fattened from about 10 to 29 months of age. Throughout the fattening period, body weights (BW) were measured regularly and the feed intake for each animal was measured daily. Residual feed intake, residual BW gain, and residual intake and BW gain were calculated as feed efficiency traits. We predicted methane production, methane yield, and methane conversion rate using previously suggested equations. These traits were calculated in the first half, latter half and the total fattening periods. After the animals were slaughtered, we obtained the carcass records. A single-trait animal model for estimating heritability and a two-trait animal model for estimating genetic correlations were applied. Heritability estimates for most of the feed efficiency and predicted methane-related traits differed largely between the first and latter halves. Feed efficiency traits in the first and latter halves had high positive genetic correlations with the same trait in the total period. There were desirable genetic correlations of feed efficiency traits with predicted methane-related and carcass traits. Our results suggest that selection for feed efficiency traits in the first or the latter half of the fattening period may improve overall feed efficiency and reduce overall methane production without any unfavorable correlated responses to carcass grade.

Sequencing of the GHRL gene by NGS reveals a multi-SNP cluster associated with feed efficiency

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Ghrelin (GHRL) is essential for growth hormone release and energy balance regulation in vertebrates. Studying gene mutations is crucial to understanding how the GHRL genotype influences economically significant traits, such as feed efficiency indicators. The study aimed to identify GHRL polymorphisms among intensively fattened lambs in Latvian sheep breeds and explore their association with feed efficiency traits. Using NGS, the full sequence of GHRL was examined in 76 lambs, supplemented by Sanger sequencing in an additional 92 lambs. A significant cluster of 52 polymorphisms exhibited perfect linkage disequilibrium. The specific SNP group showed a statistically significant association with feed efficiency (FE), feed conversion ratio (FCR), Residual feed intake (RFI), and Residual intake and body weight gain (RIG). Rare allele genotypes were favoured for enhancing specific traits. Intensively fattened lambs with rare homozygous genotypes for all investigated SNPs, on average, demonstrated a 75.71 g higher live weight gain per 1kg of dry material intake (DMI) and required 1.46 kg less DMI for the same weight gain than frequent allele homozygous lambs. Although requiring 0.41 kg less feed than expected lambs with the rare allele homozygous genotype had a 3-point higher RIG than those with heterozygous or frequent allele homozygous genotypes. Our findings underscore the significance of GHRL SNPs in intensive lamb fattening, suggesting their potential use in breeding to enhance Latvian sheep breeds and overall economic yield. Acknowledgements. The study was funded by the LZP-2021/1-0489 project.

On the use of mid-infrared spectra and physiological measures for the characterization of dairy cattle thermotolerance

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In livestock farming, efforts to combat the consequences of climate change (CC) focus mainly on mitigating the impact of livestock on climate and reducing the impact of CC on animals. For the latter, breeding tools are presented as a promising strategy. One of the main challenges is the need to define selection criteria that allows us to characterize individual thermotolerance, thus allowing us to select for more thermotolerant animals. The present study explored the use of mid-infrared (MIR) spectra from routine milk recording schemes to discriminate heat stress vs. thermoneutral samples and to classify heat tolerant vs. heat susceptible cows. MIR data from monthly milk recordings was available for 620 dairy cows. In addition, respiration rate (RR) was measured during a heat wave on a sample of 230 cows with MIR data available. Those cows were categorized as heat tolerant or heat susceptible based on RR. A PLS-DA model was used to predict group membership using the MIR data. The prediction of the heat load group of the individuals based on MIR data resulted in relatively good accuracy when considering the average daily temperature as a measure of heat load. However, the prediction accuracy was low when attempting to characterize the thermotolerance group based on RR. To address the identification of patterns of response to heat stress in individuals according to their degree of thermotolerance, more data (MIR data and RR evaluations) are needed to characterize the degree of individual thermotolerance in a more precise way. This work has received funding from the European Union's Horizon Europe research and innovation programme under the grant agreement No 01059609 (Re-Livestock project)

A single-step genomic evaluation of maturing rate index in French beef breeds

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In beef farming, one way to maximize productivity while limiting environmental impact is to select animals that develop more quickly. The maturing rate index, which tracks the proportion of adult weight the individual has reached over time, can be used to measure this precocity. A preliminary study revealed that the maturing rate index, estimated using the Brody equation, was moderately heritable. In this study, we assessed implementing genomic selection for this trait in Charolais, Limousin, Blonde d'Aquitaine, Parthenaise, and Aubrac breeds. Weight records from birth to post-weaning from commercial farms and slaughter weights from French national databases were used. For more reliability, only females slaughtered after 4 years of age with at least 4 recorded weights were kept. The final dataset included data on over 460,000 Charolais, 270,000 Limousin, 93,000 Blonde d'Aquitaine, 38,000 Parthenaise, and 7,000 Aubrac females, with respectively 8,000, 2,000, 4,000, 1,000, and 600 genotyped animals. An univariate genetic evaluation was performed using a single-step model with the HSSGBLUP software in order to estimate the breeding values for the maturing rate index. Across the five breeds, the heritability of maturing rate index ranged from 0.22 to 0.25. The genetic evaluation revealed that gaining one standard deviation of the precocity of development genomic index equated to a maturing rate index increase of 9.86% and a reduction of 1.3 months to reach half of adult weight. Routine implementation of this evaluation is anticipated in the near future, offering the opportunity to select the most precocious animals in beef farming. Furthermore, investigating correlations with other key traits targeted for selection is recommended.

Genetic relationship between young bulls' feed intake and GHG emissions measured on cows' heat tolerance measures in Italian Holstein cows

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Aim of this study was to analyse the breeding values for feed efficiency and greenhouse gas (GHG) emissions in relation to breeding values for heat tolerance (IHT) in young Italian Holstein bulls. For this study young Italian Holstein bulls tested at the Center of Italian National Breeders Association for the Holstein, Brown and Jersey dairy cattle breeds (ANAFIBJ) have been used. Feed intake measures were obtained using the Roughage Intake Control system (RIC) while GHG emission traits were derived from measures taken using the GreenFeed (C-Lock Inc.). Phenotypic data was provided for on 218 Holstein bulls between the age of 171 and 541 days. All bulls were genotyped using various SNP chips resulting in 69,127 SNP after imputation and editing. The Italian Heat Tolerance Index (IHT) for daily milk production was estimated using a genotypeXenvironment approach and the temperature-humidity index (THI) considered the interaction with the animal. A single-trait repeatability linear animal model, considering a pre-estimated heritability of 0.16 was used. Data were available for 159 to 202 days of age young bulls. Spearman rank correlation was used to relate the genomic breeding values between the two set of traits. Results showed a weak favourable relationship between IHT and CO₂ emissions (-0.17) and CH₄ emissions (-0.08). Results showed a negligible relationship between IHT and feed intake measures, with all correlations being close to zero.

Identification of functional SNPs linked to feed efficiency in Canadian Beef cattle

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Beef production contributes to approximately 2.4% of Canada's total Greenhouse Gas emissions. Improving beef cattle feed efficiency (FE) may lead to improved energy partitioning and resource use, thereby improving sustainability. Our objective was to identify novel functional SNP markers linked to FE in Canadian beef cattle using RNA-Sequencing (RNA-Seq) of rumen tissue collected from 48 beef cattle [n=16 Angus, n=16 Charolais, n=16 Kinsella (Composite Hybrid including Angus, Charolais, Galloway, Hereford, Holstein, Brown Swiss, and Simmental)] selected for extreme FE phenotypes. In total, 11 key regulatory genes (MYH1, MYL2, MYLPP, TNNC2, EIF4B, RHOD, TCEANC, CKM, ENSBTAG00000040518, SERPINB2, and USP43) were significantly differential expressed (DE) between extreme Residual Feed Intake (RFI) groups (low-RFI n=8, high-RFI n=8 per breed) using CLC Genomics Workbench (FDR<0.05; |FC|>2). Using an optimized RNA-Seq variant calling pipeline using STAR and BCFtools, a total of [total (unique to low-RFI, unique to high-RFI)] 75 (36, 39), 78 (42, 36), and 53 (35, 18) uniquely fixed functional SNPs were located within coding regions of these 11 functional candidate genes, in low- and high-RFI animals in the Angus, Charolais, and Kinsella breeds, respectively. Considering all functional SNPs uniquely identified in low- or high-RFI groups for all breed comparisons, the majority of SNPs were identified in MYH1, EIF4B, and SERPINB2 genes, which function together in highly metabolically demanding biological processes (P<0.05) related to muscle contraction, system, and filament function processes. Using RNA-Seq to identify key regulatory genes and associated functional SNPs linked to FE may uncover important genetic markers that influence the regulation of FE in beef cattle.

Is methane emission the same trait genetically in young bulls and lactating dairy cows?

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It is of interest to examine whether methane (CH₄) emission is the same trait genetically in young bulls and in lactating dairy cows. The aim was therefore to estimate the genetic correlation between CH₄ emissions for Norwegian Red young bulls and cows in lactation. Measures of CH₄ from GreenFeed (GF) was available from Geno's test station for young bulls and from 15 GF units installed in commercial dairy herds. Data from the years 2020 – 2023 were included and after edits we had a dataset with 771 989 GF visits from 1 482 cows, and from the test station we had 112 071 GF visits from 244 young bulls, measured at 11-12 months age. Each young bull had on average 40 days with CH₄ data. Each visit at the GF provides an estimate of the animals daily CH₄ emission. The traits analyzed were gram CH₄ per cow or bull per day, computed as the average of the individual visits each day. The mean (standard deviation) of daily CH₄ for cows and young bulls were 406 (108) and 222 (46) gram CH₄, respectively. A bivariate linear animal repeatability model was used to estimate (co)variance components. The estimated heritability (standard error (s.e.)) of CH₄ emissions was 0.39 (0.04) for cows and 0.49 (0.15) for young bulls, and the estimated genetic correlation (s.e.) between the two traits was 0.63 (0.22). The large s.e. reflects that the genetic correlation was estimated based on information from few animals and should therefore be interpreted with caution. The results suggests that phenotyping future young AI bulls is valuable for genetic evaluation of methane emission in Norwegian Red, even if CH₄ emission is not exactly the same trait genetically in young bulls and lactating cows.

Genetic evaluation for feed efficiency in German Holsteins

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Genomic evaluation was developed for feed efficiency for German Holsteins and the first official release is April 2024. The data set consists of 327,408 weekly records of dry matter intake (DMI), body weight (BW) and energy-corrected milk (ECM); collected from 13,914 cows over the first three parties from six countries as part of the resilient dairy genome project. Each parity was divided into four equal lactation stages and then a single-trait three-parities animal model was utilized to estimate variance components. These estimates were used to derive the random regression coefficients using the covariance function approach. Subsequently, a single-step random regression model was implemented. The herd-test-week, regression on inbreeding and the calving age class by week of lactation were the fixed effects. Random effects included permanent environmental and additive genetic animal. The averages of the first parity weekly heritability estimates were 0.19, 0.30 and 0.48 for DMI, ECM and BW, respectively. The lowest genetic correlations of DMI between lactation weeks were 0.65, 0.52 and 0.70 for parities 1 to 3, respectively. The genetic correlations of DMI between the three parities were higher than 0.79. Body weight change (BWC) estimated breeding values (EBV) were derived from the BW EBV. Feed saved (FS), which is EBV for feed efficiency (DMI in kg), was estimated as $ECM\ EBV \times 0.4 + BWC\ EBV \times 4.5 - DMI\ EBV$. The genetic correlations of FS with the milk production index RZM and other main indices in the total merit index were around zero. The FS genetic standard deviation was 247 kg per 305 days in milk, which is roughly 3.5% of DMI. Breeding for feed-efficient cows will enhance the profitability and sustainability of the dairy cattle industry in Germany.

Investigating Methane and Carbon Dioxide Emissions in Hanwoo Steers: Implication of Age and Genetic Traits

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This study explores the effects of age and genetic traits on methane (CH₄) and carbon dioxide (CO₂) emissions in Hanwoo steers, a breed of cattle native to Korea. Forty-eight steers were categorized by age (10-month-old vs. 13-month-old) and genetic traits (Q=quality grade vs. G=yield grade), resulting in four groups: 10Q (BW= 293 ± 29.50 kg), 10G (BW= 321 ± 27.45 kg), 13Q (400 ± 33.98 kg) and 13G (424 ± 38.82 kg). Steers were fed a 60:40 forage-to-concentrate ratio adjusted for their age difference. Greenhouse gas (GHG) emissions, specifically CH₄ and CO₂, from Hanwoo steers were monitored using the GreenFeed unit. Results demonstrated significant age-related effects on CO₂ emission (g/d), CH₄/CO₂ (0.06 CH₄L/d), and CH₄/CO₂ (0.10 CH₄L/d) (P<0.0001, for all). Age and genetic traits influenced dry matter intake, and metabolic weight (P<0.01, for both). Spearman correlation analysis revealed positive correlations between the group and BW (r=0.84, P<0.0001) and CO₂ emission (g/d) (r=0.87, P<0.0001). Moreover, CH₄ emission (g/d) correlated strongly with CH₄ yield (g/d/kg) (r=0.93, P<0.0001) and CH₄ intensity (g/d/kg BW^{0.75}) (r=0.93, P<0.0001), indicating mutual influence. These findings highlight the importance of understanding age-genetic trait interactions in GHG emissions from Hanwoo steers for developing strategies to mitigate livestock production's environmental impact while enhancing animal productivity and welfare.

Impact of Temperature-Humidity Index on Milk Production in Crossbred Dairy Cows

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The aim of this study was to evaluate the influence of thermal stress, estimated through the environmental Temperature and Humidity Index, on the productivity in triple-cross breed primiparous cows in Argentina. Retrospective data from 1635 heads spanning the years 2009 to 2020 were analyzed. Three different breed groups resulting from Holstein x Jersey crosses, incorporating semen from Brown Swiss, Guernsey, and Montbeliarde breeds. The THI 60-days in before its birth (bb) and after its birth (ab) during. A day with thermal stress was defined as one with at least 7 continuous hours of THI_h ≥ 72 during the spring and summer seasons. Stressed was defined if it experienced at least 10 consecutive days of DTS. The dependent variables investigated were milk yield (MY305) and the age at first calving of the cows (C_AFC). The latter pattern was categorized into three clusters (cat I: < 24; cat II: 24 < cat II < 30; cat III: > 30, months). To analyze the impact of heat stress on MY305, a mixed linear model was applied. Fixed effects included C_AFC, BG, Stress_ab, Stress_bb, their two-way interactions and Stress_bb- Stress_ab- RG interaction, while year and month of cow's calving were considered random effects. Significant differences were observed in MY305 for C_AFC (p=0.00179), RG x C_AFC interaction (p=0.00016), and Stress_ab x C_AFC interaction (p=0.0242297). These findings highlight substantial variations between Cat_I - Stress_ab (4074 ± 352) and Cat_I - Non Stress_ab (5239 ± 138) in thermal stress. In conclusion, the impact Stress ab on the milk production of different dairy genotypes.

Modelling the environmental and genetic effects of climate conditions on the growth of a local cattle breed
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Managing heat stress in cattle is a major challenge in the current era of climate change. The aim of this study is to evaluate the effect of Temperature Humidity Index (THI) in relation to body weight (BW) and average daily gain (ADG), and exploring potential genotype-by-environment (GxE) interactions in the local Italian Rendena breed. Phenotypic data from performance tests (PT) of 1792 bulls conducted since 1985 were used. Individual ADGs were computed from monthly weightings, resulting in a final dataset of approximately 10 observations per young bull of ADG and BW. Moreover the overall ADG of PT was considered. Climatic data from the specific PT area were extracted from the NASA and OPEN-METEO services databases. From this data the average THI, the average maximum THI and the mean THI for specific risk time slots (10AM-6PM, 12PM-5PM and 12PM-3PM) were computed and linked to each monthly ADG and BW, and incorporated into the model. In terms of the models, in addition to genetic effects, we included THI, cow calving order, performance period, season, test group and random individual. THI was also incorporated as a random slope within random regression models. Then, the inclusion of pedigree information allowed to estimate additive variance and possible GxE interactions for the traits under investigation. A significant negative influence on traits was observed along with the increasing of THI, or its risk levels if THI is considered as factor. The heritability for overall ADG and BW was respectively around 0.3 and 0.19 on average. We also identified a small but existing effect of GxE on the total genetic variance. Given the importance of the traits in defining the productive capacity in cattle, a proper knowledge of the effects of climate variables as THI is a key to obtain an effective genetic improvement. The study highlighted the importance of evaluating inclusion of THI in routine estimations of ADG. The Authors acknowledge the National Breeders Association for Rendena Breed (ANARE) for data and the PNRR project DM118/2023 for funding.

Exploring potential GxE interactions in the lifetime performance of suckling ewes

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Prolific ewes produce more lambs per time and are often preferred by breeders. Yet, in grazing systems, those ewes might not be able to cope with feed resource limitations that are increasing in frequency, severity and duration with climate change. To explore the effects of possible feed constraints at different prolificacy potentials, we simulated the ewe's lifetime lamb production in two contrasting pasture scenarios using a mechanistic model of energy acquisition and allocation between functions. Each ewe genotype was defined by several parameters controlling energy acquisition and allocation, including the prolificacy potential. In the favorable scenario, grass regrowth was maintained high during summer, whereas it was strongly impacted by drought in the unfavorable scenario. In both scenarios, ewes with higher prolificacy potential weaned on average more lambs over their life. Most ewes living long also weaned more lambs. There was no clear trade-off between ewe longevity and prolificacy. In very prolific ewes, the number of lambs weaned during lifetime was more strongly correlated between environments ($r = 0.69$ to 0.78) compared to less prolific ewes ($r = 0.26$ to 0.43). Those correlations suggest that ranking of ewes in one scenario did not match well the ranking in the other, especially at lower prolificacy potential. However, a high prolificacy potential was not enough to ensure a high and robust lifetime performance. The best prolific ewes in the favorable scenario showed a wide range of performance in the unfavorable scenario. Generally, the best ewe genotypes combined a high prolificacy potential with a balanced energy acquisition and allocation strategy that allowed them to both maintain their body reserves and to mitigate the higher lamb mortality observed in large litters. Although our model suggests the existence of GxE interactions, further analyses integrating real data will be needed to assess the genetic component of energy allocation strategies.

Genetic diversity of South American Criollo cattle adapted to a wide range of environmental conditions
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In South America, cattle breeds were introduced from the Iberian Peninsula soon after the New World discovery. Over centuries of human colonization, these cattle dispersed throughout the continent and adapted to diverse climates and husbandry practices. Later on, other taurine breeds imported from Europe, and indicine breeds from India and Pakistan enriched the genetic landscape of the continent. The original Iberian are still reared in extreme environments. We explored the molecular diversity and climate adaptation of 979 criollos from 18 breeds across eight countries and 10 agro-ecological zones. A total of 24,831 SNPs were used for assessing genetic diversity, population structure, selection signature and climate adaptation of criollo cattle. Results revealed notable heterogeneity within populations, highlighted significant gene flow, admixture events from various source populations and genomic region having adaptive value.

Simplified workflow for faster genotyping-by-sequencing using partial combinatorial dual barcodes and optimized analysis pipeline

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Targeted Genotyping by Sequencing (GBS) is a robust and cost-effective method for marker-assisted breeding and selection. Targeted GBS provides a scalable workflow, allowing for thousands of markers to be analyzed in up to thousands of samples per day. As GBS use expands, demand increases for higher throughput, faster turnaround, and increased multiplexing capability in GBS workflows. To achieve higher throughput with less material and greater ease of use, we propose a new library prep workflow using partial combinatorial barcodes for use with the Ion GeneStudio™ S5 System that not only increases available barcodes with less material, but also achieves complete, normalized libraries for a minimum of 3072 samples in less than a day. Additionally, we propose an analysis workflow optimized for ~6000 samples to be sequenced and genotyped in less than 24 hours using a single Ion GeneStudio™ S5 System. These optimizations enable completion of genotyping of ~30,000 samples within a 5-day week. Here we report early testing results showing high quality results achieved using this novel workflow and analysis pipeline.

Assessment and distribution of Runs of Homozygosity and their relation to water stress in the Rasa aragonesa sheep breed

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Heat stress and scarcity of drinking water for animals is a characteristic feature in the semi-arid region where the Rasa Aragonesa sheep breed is reared, especially during summer months with high temperatures. The aim of this study was to estimate the number, length, and frequency of runs of homozygosity (ROH) in Rasa Aragonesa ewes and identified genomic regions with a high ROH frequency using the 680k Illumina AgResearch Sheep HD chip. Moreover, we sought to identify ROHs associated to water restriction and heat stress in animals with different response to these stresses. In total, 201 ewes were subjected to total water restriction for 5 days, experiencing heat stress conditions during 55% of the experiment. Blood samples were collected at day 0 and 5 for haematological and metabolite measurements. Wool samples were taken at day 0 and 4 weeks later (28d). Hierarchical clustering was performed based on the variation of the blood and wool traits. The PLINK software was used for quality control (QC), while detectRuns package was used to detect ROH with the sliding-window option and establishing 1 Mb as the minimum length of a ROH. Hierarchical clustering predicted 4 clusters: 1) fat mobilization ewes (n= 56), 2) high-stressed ewes (n= 9), 3) low-stressed ewes (n= 106), 4) stressed ewes (n= 31). Furthermore, two main clusters were found based on low (cluster 1 – 2) or high dehydration (cluster 3 – 4). After genotype QC, 531,212 SNPs were retained. A total of 2130 ROH exceeding 1 Mb were detected in the whole data, with an average number of ROHs and length in each animal of 10.6 and 4.13 Mb, respectively. Most of the ROH were less than 5 Mb (70%). The inbreeding coefficient (FROH) was 0.025. Cluster 3 and 4 showed the highest and lowest average number of ROHs with 11.18 and 8.22 (with an average length of 7.07 and 3.60 Mb), respectively. Nine and 22 regions were found to contain ROH hotspots when analysing the whole dataset, and the high and low dehydration clusters, respectively. Candidate genes that could be related to water stress were identified within these genome regions.

Session 2

Theatre 1

Economic values for organic dairy cattle production systems in Denmark

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Denmark is among the world-leading countries in organic dairy production. However, all breeding material for dairy cows originates from conventional dairy production, where organic cows are selected based on the Nordic Total Merit index. This may be a problem since organic production differs from conventional production in e.g. use of antibiotics, feeding, usage of reproductive technologies such as multiple ovulation and embryo transfer, and prices for feed and milk. If we use cows and bulls selected based on the index for conventional production, we may therefore not obtain the optimal genetic gain from an organic point of view. Implementation of a specific organic breeding goal will also make it possible to market organic products with an organic breeding profile. The aim of this study was to define a breeding goal based on economic models and preferences of consumers, dairy companies, and farmers. In this first part of this study, we defined production systems and traits for organic dairy production. Economic values were derived using the SimHerd bio-economic model, which models a dairy cattle production system in detail. We used data from the Danish Red dairy breed since this breed is already aiming for being suited for organic production in the future. We expect that the organic breeding goal will have more emphasis on climate impact and animal welfare compared to the conventional current one.

Modelling the transition from conventional to genomic selection in German Merino sheep breeding by stochastic simulations

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Selection in the German Merino sheep breeding program is currently based on a pedigree BLUP breeding value estimation (BVE). A simulation study was conducted using the R-package MoBPS to simulate the German Merino breeding scheme. Different selection strategies based on a single-step GBLUP BVE were compared to a reference scenario with pedigree-based BVE. Real genotype data of German Merino was used to create a population specific simulation. A health and a production trait were simulated with selection based on a selection index with equal weights for the two traits. Scenarios were simulated in which a preselected share of the top 25% (top 50%) of male lambs was genotyped. In a further scenario, all male lambs were genotyped. Genetic gain increased with increasing numbers of available genotypes, however, marginal gains decreased. Genotyping the top male 25% already led to +13% in genetic gain, whereas genotyping all males led to an increase of +23% compared to the reference scenario. Further scenarios were simulated in which, additionally to all male, the top 25% (top 50%) or all female lambs were genotyped. Genotyping female lambs significantly increased genetic gain (+31% to 33%) but without significant differences between the proportions of genotyped females. Results confirmed the potential of genomic selection for German Merino sheep and showed that even a certain proportion of genotyped male lambs can already lead to significant improvement. This study might be used as a base for discussions on how to implement genomic selection into the breeding program.

Session 2

Theatre 3

Only 100% to share - breeding objectives revisited for improved sustainability and animal welfare

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Performance traits have long been the focus of livestock breeding programs. Genetic selection and management improvements have resulted in remarkable phenotypic and genetic gains in many populations worldwide. However, in order to overcome the one-sided breeding for performance with its increasingly negative effects on fitness, more and more functional traits have found their way into breeding goals. For example, in many dairy and dual-purpose cattle populations, total merit indices combine a large number of different traits, including those related to production as well as to fertility, health, survival or feed efficiency to name a few. Despite the typically low heritabilities and mainly negative genetic correlations with production traits of the latter, genetic gains can also be achieved for these traits if they are given appropriate weight in the breeding objectives. An increasing number of auxiliary traits, e.g. from automation, and the use of genomic evaluation are making this even more feasible. To meet the demands for improved animal welfare and sustainability, a larger number of traits or trait complexes will likely need to be considered in future breeding objectives. Greenhouse gas emissions to reduce environmental impact and resilience to climate change are certainly among them. They are linked to sustainability, efficiency, animal welfare and potentially product quality. The choice of additional traits, but also their weighting in the indices, is more challenging if at least a stable genetic trend is to be expected for each of them. Moreover, different stakeholder groups may have different preferences. How do we deal with this dilemma?

Stratified mating quota: a novel approach to effectively reduce the inbreeding rate

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Maintaining genetic diversity is important to keep a population (genetically) healthy. To do so, the FAO recommends to limit the inbreeding rate to below 1% per generation, and preferably below 0.5%. However, many breeds show high inbreeding rates, due to small population sizes and popular sire effects. In such breeds, strategies are needed to reduce the inbreeding rate in a practically feasible way. We stochastically simulated the effect of breeding strategies in MoBPS software, using pedigree and/or genotype data of the Friesian horse population. We used the Friesian horse as a case study, which is a breed that suffers from consequences of too high inbreeding rates, has a high pedigree depth and – since recently – has genotype data available. Here, we suggest a novel approach, stratified mating quota, where each sire was assigned a mating quorum (of max 60, 80, 100, or 120 offspring per sire per year) based on its mean kinship. Among the simulated breeding strategies, stratified mating quota was the most promising and feasible approach to further limit the inbreeding rate below 0.5%. For the calculation of mean kinship, we considered the impact of genotype data (70K SNP chip) on the assigned mating quota. We calculated genomic relationships between breeding sires (n=72) and foals born in 2023 (n=2,886) and compared this to their pedigree-based relationships. By applying stratified mating quota in the Friesian horse in practice, and by promoting this approach for other breeds, we aim to further reduce the inbreeding rate in (genetically) small populations.

Session 2

Theatre 5

Using alternative relationship matrices for genomic prediction and managing genetic diversity

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Modern breeding should make fast genetic progress to feed a growing world population, and also maintain the genetic diversity of the selective populations to meet future economic, environmental and societal demands. Optimum contribution selection (OCS) is considering both. It aims to maximize the breeding efforts, whilst constraining the rate of inbreeding. Before the genome era, only the numerator relationship matrix (NRM) from pedigree was available for inbreeding management. Genomic information has provided more measures of genetic relationships. The objectives of this study are to compare genetic improvements and diversity management in a long-term breeding program, using various relationship matrices as tools. Stochastic simulation software was developed to mimic a cattle population. Visible chip SNP, as well as invisible QTL and reference SNP were sampled to form a base population. NRM, the genomic relationship matrix (GRM), and IBD relationship matrix (IRM) were used to estimate breeding values, and/or as relationship constraints for optimum contribution selections. We conclude that using IRM for diversity management performed best in maintaining population diversity. IRM diversity management yielded least inbreeding, least loss of alleles across the genome and, most genetic and genic variance, and most potential for future breeding. The final allele reduced frequency of low minor allele frequency loci, making them less prone to fixation by genetic drift. The diversity maintained also provides more resources for later genetic progress. OCS schemes generate >25% more genetic progress than non-OCS schemes over 20 generations of selection in our simulations.

Study of genetic progress in the context of disconnection between two originally connected populations

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Genetic progress of breeding programs is highly dependent on the size of reference populations and the relatedness between reference populations and selection candidates. Thus, for selection programs of small size, segregation of the population into several subpopulations could have significant consequences on selection efficiency. Organic farming may lead to organic farms breaking away from the conventional scheme to form a distinct breeding scheme. The aim of our study is to investigate, on the basis of stochastic simulations, the impact of such separation. We simulated a dairy scheme with 5,400 females mated to 90 males and a trait of heritability 0.30. After several cycles of pedigree-based and then genomic selection, the population split into two subpopulations, still under a common evaluation, with different ratios: 50/50, 60/40 and 80/20. We studied the evolution of F_{st} , accuracy of predictions and genetic progress over generations. We observe a faster genetic divergence in the case of unbalanced separation (F_{st} from 0.01 to 0.05 and to 0.08 for the scenario 50/50 and 80/20 respectively). Splitting the population has a negative effect on the genetic gain for the smallest population compared to the population without separation: 0.321 (0.044) against 0.335 (0.020) and 0.316 (0.030) against 0.327 (0.020) respectively for the scenarios 80/20 and 60/40. We do not find any difference in the case of balanced splitting.

Session 2

Theatre 7

A comparison between phenotypic and genotypic strategies of crossbred individuals in a crossbred scheme of laying chickens

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The laying hen industry significantly contributes to the global food supply. The major companies in the sector have many pure lines that cross to be able to adapt to the needs of the market. However, information from commercial crosses might be underutilized in breeding programs. This study aims to assess the extent to which incorporating crossbred information can enhance evaluations and determine the optimal phenotyping and genotyping scenario. To achieve this, a simulation of a standard layer breeding scheme was conducted using the AlphaSimR program. Genetic evaluations of this scheme were then carried out using the BLUPF90+ software tool. Correlations between actual breeding values and those estimated were analyzed for various genotyping and phenotyping scenarios for crossbred hens. The results revealed a remarkable increase in correlation when hens were individually phenotyped and genotyped. However, the challenges associated with the collection of this information, particularly phenotyping, make this scenario difficult to implement nowadays. Alternatively, acquiring individual genotype and cage means, which is more cost-effective and offers improved precision to a lesser extent, could be a viable option. Future perspectives of this study may include exploring other traits with different heritabilities and correlations, conducting a cost/benefit analysis by mimicking a real-world situation, and investigating response to selection over multiple generations rather than precision alone.

Selection for antagonistic traits must balance diversity in terms of genetic correlation

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Genetic parameters have long been considered as static in genetic evaluations. The principle of ignorable selection ensures that multi-generational predicted breeding values are as accurate as possible, given (co)variance parameters estimated at a base population. It is widely known that selection leads to a loss of genetic diversity, i.e. a decrease in genetic variances, a phenomenon well known as Bulmer effect. However, the Bulmer effect dissipates after a few generations, and genetic variances are rather stabilized, being most changes then due to random drift. While losses of genetic diversity due to losses in genetic variances has been well studied by quantitative geneticists, consequences of changes in genetic correlations on genetic diversity are little explored. The influence of genetic correlations on diversity is of particular concern when dealing with negatively correlated traits. While selection for two antagonistic traits ensures genetic gains for both, in a medium-to-long-term such selection may favor individuals at the border of the bivariate distribution of the negatively correlated breeding values, drastically reducing genetic diversity with respect to their capacity of trade-off regulation. The medium-to-long-term consequence of this loss in genetic diversity is that mutual genetic gains for both traits of interest may no longer be a possible option in a breeding program. This risk of loss of genetic diversity can be controlled by adequately balancing the breeding goals for the traits involved, and with a simulation study we observed that this balance is strongly dependent on the traits h^2 . This project has received funding from the European Union's Horizon 2020 Programme for Research & Innovation under grant agreement n°101000226.

Session 2

Theatre 9

New insight in genomic inbreeding assessment in domestic animals

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The assessment of inbreeding is one of the most important indicators of genetic variability. The common method for calculating genomic inbreeding is Runs Of Homozygosity (ROH) based inbreeding coefficient (FROH). Despite the potential of ROHs to provide a regional approach to inbreeding assessment, this aspect remains largely unexplored, as all studies have focused solely on entire chromosomes or the entire genome. Thus, in this study, we analysed regional inbreeding variation on each chromosome to see exactly where inbreeding is more pronounced and in which regions there are large differences between the sexes. To do this, we presented three case studies for three different domestic species: Cattle (79 females and 33 males; Illumina BovineHD SNP Chip), Dogs (128 females and 148 males; Illumina CanineHD BeadChip) and Sheep (101 females and 102 males; Illumina OvineHD BeadChip). For each species, regional FROH were estimated separately in males and females using SVS and RZooROH software. Regions of large inbreeding differences were detected in all three cases, with much greater variability observed on the X chromosome than on the autosomes. Our results suggest that greater precision can be achieved in monitoring inbreeding at the regional level, further contributing to breeding management, particularly in terms of tracking inbreeding depression for various important traits.

Prediction of hidden individual inbreeding depression load in French dairy sheep

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As well as lethal mutations, other (usually unknown) recessive conditions that are not lethal are carried by individuals, in what is known as load. Individual variability exists in this load of unknown deleterious alleles. It is possible to define and estimate a polygenic individual Inbreeding Depression Load (IDL). Each individual possesses parts of inbreeding coming potentially from different ancestors. Using these parts, a linear model then predicts the IDL of the individuals, which is an additive trait that is expressed only in inbred individuals. The objective of this work was to estimate the genetic variance of IDL for milk yield in 3 dairy sheep breeds: Basco-Béarnaise (BB), Manech Tête Noire (MTN) and Manech Tête Rousse (MTR). A total of 658,731, 541,180 and 2,168,454 records of milk yield from 178,123, 151,863 and 596,586 females of BB, MTN and MTR, respectively, were included. Pedigrees included 190,276, 166,028 and 633,655 animals in BB, MTN and MTR, respectively, born between 1985 and 2021. Estimates of IDL genetic variance clearly indicate variability of IDL for milk yield among ancestor families in MTN and MTR. The existence of IDL genetic variance was not conclusive in BB. The correlation between additive genetic value and the IDL was -0.01. Results confirm the presence of heterogeneity in the IDL among individuals. Animals with less load could be selected to avoid inbreeding depression in future generations and undesirable matings can be discarded.

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Genotype by environment interaction and response to selection for milk production traits in Lacaune sheep

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The objective was two-fold (i) to investigate genotype by environment (G×E) interaction for milk yield and composition in purebred Lacaune sheep reared in Greece and France and (ii) to compare potential genetic gain from selection across and within countries. A total of 1,658 Lacaune ewes from four intensive farms in Northern Greece and 4,859 Lacaune ewes from 186 semi-extensive farms in Southern France were used. Ewes in the two countries were daughters or granddaughters of the same rams. Individual ewe daily milk yield was recorded monthly and milk samples were collected to assess chemical composition. Total milk yield and milk component (fat and protein) content were calculated. (Co)variance components were estimated using ASReml software. Results showed a strong genetic correlation for milk yield (0.86±0.13) and protein content (0.88±0.12) between the two countries suggesting no presence of G×E interaction. Regarding fat content, a moderate correlation was found (0.59±0.21) indicating some degree of sire re-ranking. Our analyses suggest that a joint genetic evaluation of Lacaune sheep in Greece and France is feasible. Simulations showed that sire selection across countries based on a joint genetic evaluation may increase genetic gain up to 68.17% and 0.16% in Greece and France, respectively, compared to selection within country. This work was funded by SMARTER Horizon 2020 project (772787).

How dual-purpose is dual-purpose? The balancing act between fattening and laying performance:
An index to describe dual-purpose poultry

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Commercial poultry production systems with specialized laying or meat strains are criticized by society and are associated with ethical and animal welfare problems. As the rearing of males of layer lines, an alternative to the resulting ban on killing of male chicks in several EU countries is resource-intensive, dual-purpose poultry can be an alternative. Especially integrated organic production systems can benefit, if the production of eggs and meat from the same genotype can be managed efficiently. For this, productive performance profiles of dual-use poultry genotypes need to be defined and evaluated. In the framework of the EU Horizon 2020 project PPILOW novel dual-purpose genotypes were evaluated under organic farming conditions. The aim of this contribution is to investigate the potential of these tested dual-purpose genotypes, evaluated as a unit in the context of poultry genotypes and how this is reflected in their economic performance. A dual-purpose index (DPI) was used, reflecting the joint performance of a dual-purpose unit as proportion of the performance of commercial hybrids and is set to 1 for dual-purpose genotypes. Higher values indicate a higher proportion of laying performance, lower values indicate a higher proportion of fattening performance. The DPI of the novel dual-purpose genotypes tested in PPILOW ranged from 1.29-1.97. The analysis serves as basis for further research to define and distinguish between single and dual-purpose types and from a business case perspective, to add value via successful on-farm implementation. This project is funded by the EU Horizon 2020 program (N°816172), carried out in collaboration with SYSAAF, Novogen, Hendrix Genetics, Aarhus University, INRAE, ITAB

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Inbreeding depression for litter size in a mice population divergently selected for environmental variability of birth weight

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The aim of this study was to estimate inbreeding depression (ID) from genomic data in the first birth litter size (LS) in two divergent mice lines selected for high variability (H-Line) and low variability (L-Line) of birth weight. A total of 1380 females of 26 selection generations were genotyped with a high-density SNP array. After applying filtering criteria, 545,656 SNPs were used for analyses. LS data of 732 L-Line and 648 of H-Line animals were used. Runs of homozygosity inbreeding (FROH) and homozygosity by descent inbreeding (FHBD) were calculated. FROH and FHBD were divided in 9 length (FROH1-2Mb to FROH≥32Mb) and age (FHBD2 to FHBD512) classes. FROH were calculated in the 19 autosomes (CHR). All the inbreeding coefficients (Fs) were standardized by the mean F of the 1st generation. The ID was calculated as the regression coefficient of LS on the different adjusted Fs. The age of the female at birth in days and the female's mother litter size were included as continuous fixed effects. PLINK v1.9, RzoRoH R package and "lm" function of R environment were used. FROH and FHBD produced significant ID in H-Line (-4.70 and -5.09) and L-Line (-4.47 and -4.55). Purge effect (P) was detected for FROH 1-2Mb (59.40) in H-Line, while ID was detected for FROH≥32Mb (-7.27) in L-Line, and P for FROH1-2Mb (44.46) and FROH4-8Mb (17.37). FHBD8 (-3.08) and FHBD256 (21.8) had significant effect in L-Line. ID was detected in CHR13 in H-Line and CHR1, CHR9 and CHR11 in L-Line. The length and the age of the ROH and HBD determined the ID and the P that were different between lines. For future research in candidate regions, CHRs 1, 9, 11 and 13 were indicated.

Genome-wide variability and selection signatures in autochthonous Spanish cattle breeds

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Morucha (MOR) and Avileña-Negra Ibérica (ANI) are two local Spanish cattle breeds appreciated for their high-quality meat production. Their subpopulations, Negras (MORB) and Cardenas (MORC) for MOR, Jarda (JAR) and Bociblanca (BOC) for ANI, can be distinguished by their coat color. Since phenotypic differences are mainly due to genome variability, the aim of this work was to investigate the genetic diversity among ANI and MOR subpopulations. With this purpose, 1352 animals (263 MORB, 629 MORC, 127 JAR and 333 for BOC) were genotyped with the Affymetrix BeadChip Microarray HD. After quality control, 33408 common SNP were retained. Genomic data were analyzed by using univariate (Wright fixation Index, FST) and multivariate approaches. For the latter, the stepwise discriminant analysis (SDA) and the canonical discriminant analysis (CDA) were applied. The highest weighted FST value (0.052) was found between BOC and MORB, that belong to the two different breeds. On the contrary, the lowest values were found between BOC and JAR (both Avileña-Negra Ibérica) and between MORC and MORB (both Morucha). The CDA was able to significantly separate MOR and ANI using only 27 SNP selected in the SDA. On the other hand, MORB and MORC, JAR and BOC, were significantly discriminated using 82 and 29 markers, respectively. Gene discovery was carried out by using the genomic regions highlighted by the two approaches. This study was able to determine the genetic difference between the analyzed breeds and among subpopulations within each breed.

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Poster 15

Networks of inbreeding in two lines of mice divergently selected for environmental birth weight variability

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The partial correlations and the information theory allow to distinguish when the association between two variables is independent from the association with a third variable. Therefore, the objective of this study was to apply partial correlations and information theory to the inbreeding coefficients (Fs) of mice selected for high variability (H-Line) and low variability (L-Line) of birth weight to detect spurious correlations. A total of 1518 individuals of 26 selection generations were genotyped with a high density SNP array. After applying filtering criteria, 545,656 SNPs were used for analyses. Pedigree inbreeding coefficient (FPED), genomic inbreeding derived from different genomic matrices (FNEJ, FL&H, FVR1, FVR2 and FYAN), from runs of homozygosity (FROH) and from homozygosity by descent probabilities (FHBD) were calculated. All the inbreeding coefficients were adjusted by the mean F of the 1st generation. Pearson's correlations and first order partial correlations were computed between all the Fs. The tolerance level was calculated using the information theory to determine if a partial correlation was significant. ENDOG v4.9, PLINK v1.9, RzoRoH R package, PCIT, Cytoscape v 3.10.1 and own code in R were used. Correlations were between 0.84 (FPED-FVR1) and 1.00 (FNEJ-FL&H) in both lines. The FVR2 correlations were not significant with FNEJ, FL&H, and FHBD. FPED, FYAN, FVR1 and FROH presented statistically significant correlations with all the rest of coefficients in both lines. The partial correlations and the information theory showed that Fs performance was the same in both selected lines and that FVR2 were less informative with respect to the other Fs.

Population dynamics of potentially harmful haplotypes: pedigree evidences

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Low-frequency haplotypes, never observed in homozygous state in a population, could be informative on potentially harmful alleles (candidate alleles). Although their identification is challenging, little is known about the dynamics of potentially lethal haplotypes. A pedigree of the highly endangered Gochu Asturcelta pig breed, including 471 individuals belonging to 51 different families with at least 5 offspring each, was genotyped using the Axiom PigHDv1 Array (658,692 SNPs). Analyses were performed on four different Cohorts (BP=10; G12=52; G23=281; G3=128), defined according to pedigree depth, and at the Whole Population (WP) level. The 4,470 Linkage Blocks identified at the BP using PLINK v1.9 gathered a total of 16,981 alleles in the WP. Up to 5,466 (32%) haplotypes were statistically considered candidate alleles, of which 3,995 (73%) had one copy only. The number of alleles and candidate alleles varied across Cohorts according to sample size. Up to 4,610 of the alleles identified in the WP (27% of the total) were present in one Cohort only. Parentage analysis identified a total of 67,742 parent-offspring incompatibilities. The number of mismatches varied according to family size. Segregation analyses informed that most potential candidate alleles appeared de novo in the pedigree. Only 17 candidate alleles were identified in the boar, sow, and paternal and maternal grandparents and were considered segregants. The occurrence of low-frequency haplotypes is likely to be caused by Allele-Drop-In events due to SNP calling errors. Only fully segregant candidate alleles should be considered potentially harmful alleles. Work partially funded by grants PID2019-103951RB/AEI/10.13039/501100011033 and PRE2020-092905 (KDA).

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Poster 17

Occurrence of haplotypes in low frequency is not explained by recombination and CNV regions only

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Linkage Blocks (LB) were defined as strong linkage disequilibrium stretches flanked by genomic areas with higher recombination rates (ρ). The identification of LB is subject to large variation depending on the methods used, the availability of a base population (BP), genetic factors, and population events. A pedigree of the highly endangered Gochu Asturcelta pig breed, including 471 individuals, was genotyped using the Axiom PigHDv1 Array (658,692 SNPs). LB were identified at the BP using PLINK v1.9. Parameter ρ was estimated in 50 kb windows using the R package FastEPRR. Relationships between LB, ρ hotspots, and 344 CNV regions were ascertained using the intersect function of Bedtools. Mean ρ estimated for the whole typed population was 2.2 ± 3.9 cM/Mba total of 212 LB (gathering 5.7% of the total number of alleles identified) overlapped with at least one window with ρ 2.5 s.d. above the mean (ρ hotspots). Furthermore, a total of 177 LB, gathering 804 different alleles (4.7% of the total number of alleles identified; 4.5 alleles per LB) overlapped with 72 different CNV regions. Only six different LB, located on SSC2, SSC6, and SSC11, gathering a total of 45 different alleles (7.5 alleles per LB), overlapped with both ρ hotspots and CNV regions. Although the role of recombination in the occurrence of new haplotypes within LB cannot be neglected, in our data the overlap of the LB identified with genomic areas potentially considered recombination hotspots is low and, therefore, recombination cannot explain by itself the excess of new alleles identified. The occurrence of Allele-Drop-in events causing the identification of new haplotypes within LB should be explored. Work partially funded by grants PID2019-103951RB/AEI/10.13039/501100011033 and PRE2020-092905 (KDA).

Genomic partitioning of inbreeding depression in Jersey cattle

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Inbreeding depression (ID), the reduction of survival and reproduction in inbred offspring, has been widely observed across almost all experimental, natural, and domesticated populations. Despite numerous research studies and experiments conducted on the mechanism of ID, its genetic and molecular basis remains elusive. Our idea intends to partition the effects of ID in the dairy cattle genome by specific annotations and investigate any significant disproportional effects of ID within certain annotations on the mean values of the traits of interest. We performed ID partitioning for four reproduction traits and three productive traits of 256,268 genotyped Jersey cattle from the U.S. CDCB (Council on Dairy Cattle Breeding) database. After quality control, we mapped 60,774 SNP chip variants onto six functional annotation categories (i.e., CDS, promoter, intron, UTR, GERP constrained elements, and remaining). We estimated the effects of ID attributed to each functional annotation category by performing a multivariate regression of phenotypes onto annotation-specific inbreeding terms and correcting relatedness with GRM. For the trait CCR (Cow Conception Rate), significant enrichment of ID was detected in Promoter with a P-value of 0.041, and the enrichment of ID (δk , defined as the ratio between the contribution of annotation k to ID over the expected contribution of annotation k to ID) of 20.250. For EFC (Early First Calving), significant enrichment of ID was detected in Promoter with a P-value of 0.019 and δk of 24.434. For milk yield, significant enrichment of ID was detected in CDS, Promoter, UTR, and intronic regions, with P-values of 0.007, 0.026, 0.004, 0.0002, and δk of 5.362, 4.273, 7.356, 1.647, respectively. It was also detected in the same genomic regions for milk fat, with P-values of 0.002, 0.005, 0.036, 0.0003, and δk of 5.768, 4.945, 5.513, 1.614, and in CDS, Promoter, and intronic regions for milk protein, with P-values of 0.0005, 0.002, 0.001, and δk of 6.404, 5.282, 1.526. Our research can help better elucidate the molecular basis of inbreeding depression in dairy cattle.

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Poster 19

Impact of the DGAT1 mutation in a Bos Taurus x Bos Indicus crossbred population in India

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The DGAT enzyme (diglyceride O-acyltransferase) plays a key role in the milk fat synthesis of cows. DGAT1 is a gene known to have a mutation in the Holstein breed (K232K / K232A) in North America and Europe, leading to a reduction of fat % with the A allele (Grisart et al., 2002). However, such a reduction is not pertinent in India, where milk from Bos Indicus is strongly preferred because of its fat content. With the support of the Bill and Melinda Gates Foundation, the Indian NGO Bharatiya Agro Industries Foundation (BAIF) has created a crossbred population of Bos Taurus x Bos indicus cattle to induce sustainable genetic progress. Our study based on about 3000 genotyped crossbred cows in 6 Indian states shows that about half of them carry one copy of the mutation and about 20% two copies, leading to a decrease of fat% of about 4%, compared to the pure Bos indicus cattle. However, this undesirable evolution can be easily reversed by selecting young genotyped crossbred AI bulls not carrying the K232A mutation.

Footprints of balancing selection in the genome of Original Valachian sheep

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The aim of this study was to detect potential footprints of balancing selection in the genome of Original Valachian sheep renowned for its ability to thrive in rugged landscapes of mountain environments and by its historical significance in the traditional Slovak agricultural landscape. In harsh environments, such as mountain regions, balancing selection could play a key role in shaping the genetic diversity of breeds by facilitating their adaptability to challenging conditions and preserving their resilience over generations. We tested the genomic data of 78 ewes and 18 rams of Original Valachian sheep genotyped by GGP Ovine50k chip. Tajima's D statistic was used to find genomic regions under balancing selection, where positive D values suggest an excess of intermediate frequency variants, possibly due to balancing selection, while negative values indicate directional selection. Tajima's D values were calculated in 250 kb sliding windows, with candidate regions defined by the top 1% of positive D values. Across autosomes, 100 potential footprints of balancing selection were identified. Autosomes 3 and 6 showed the highest number of signals with average D values of 3.70 and 3.72, respectively. Subsequent gene enrichment analysis showed that protein-coding genes inside detected regions on autosomes 3 and 6 are included in various KEGG pathways, such as Toll-like receptor signalling pathway. This research was supported by the projects APVV-20-0161, APVV-17-0060, DS-FR-22-0016 and 8X23019.

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Poster 21

How directional artificial selection has shaped the genome of Italian heavy pig breeds

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The breeding programs of the Italian heavy pig breeds (Italian Duroc, Italian Landrace and Italian Large White) started in 1990, having as main objectives the needs of the Italian dry-cured ham protected designation of origin (PDO) value chains. This directional selection was based on several traits that substantially improved performance, reproduction and carcass traits of the three breeds while maintaining the meat quality for seasoning. Because of the selection, the genetic background of the three breeds diverged from the cosmopolitan Duroc, Landrace and Large White breeds used in other production systems. In this study, we monitored how this selection acted at the genomic level on the Italian heavy pig breeds. About 13,000 pigs of the three Italian breeds, born over almost four decades, were genotyped with a high-density SNP array. Over time changes of single marker and haplotype frequencies were evaluated, using a logistic regression model. A total of about 350-500 haplotypes in the three breeds significantly changed their frequency providing a picture of the genomic regions that were indirectly modified by the directional artificial selection programs operating over this period. Acknowledgements: Funded by the Programma di Sviluppo Rurale Nazionale (PSRN) SUIS-2, co-funded by the European Agricultural Fund for Rural Development of the European Union and by the MASAF.

Influence of inbreeding on reproduction traits in Swiss pig breeds

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In the present study, the relation between inbreeding coefficient and reproductive traits of proportion of piglets below 1 kg (PB1, n=28'891), piglets survival during suckling (PS, n=35'487), number born alive (NBA, n=36'477), proportion of stillborn piglets (PSB, n=97'062) and days weaning to oestrus at first litter (WO, n=27'834) were studied in Swiss pig breeds. The data were recorded over 17 years from 2002 to 2018 in Swiss Large white (SLW) and Swiss Landrace (SLR) pure lines as well as F1-crossbreds sows (SLW x SLR). The relation between inbreeding and reproductive traits PB1, PS, NBA and WO has been investigated by a linear mixed model. The proportion of stillborn piglets (PSB) was modeled using a mixed model and binomial distribution. For the traits PB1, PSB and NBA, the fixed effects of blood proportion, age at farrowing, type of insemination, litter month, litter year, piglet breed group, inbreeding coefficients of sow, boar and piglet, and random effects of boar, sow and farm were included in the model. For traits PS and WO, the effects of number of piglets suckled and suckling duration are additionally considered, and the type of insemination is omitted. Since only first litters are considered for the WO trait, there were no random effects for boars and sows in the model. The number of piglets born alive decreased as sow inbreeding increased (-2.35 ± 1.01 , p-value=0.02). For the trait PB1, none of the three inbreeding coefficients was significant. However, when inbreeding is divided into categories, the percentage of underweight piglets was significantly lower in inbred piglets than in non-inbred piglets. The piglet survival during suckling was significantly influenced by piglet inbreeding (-0.09 ± 0.03 , p-value=0.00). The weaning interval increased by 5.47 days when sow inbreeding increased by 1% (p-value=0.026). Regarding the percentage of stillborn piglets, a significant effect of piglet inbreeding was observed (odds ratio = 2.26; p-value=0.00). The results are consistent with other studies and show that the inbreeding of sow and piglet influence the reproductive traits.

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Poster 23

Rapid, low-input, targeted NGS workflow for DNA methylation in dogs

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DNA methylation is an epigenetic process involved in the regulation of mammalian gene expression. Methylation primarily occurs by the modification of cytosine bases in CpG (cytosine-phospho-guanine) dinucleotides. In canine biology, CpG markers have been associated with cancers, disease processes, development genes, and in age determination. We have developed a fast, flexible next-generation sequencing workflow for characterizing and quantitating methylation of specific markers in the canine genome. To test feasibility of our workflow we identified 100 CpG markers associated with canine aging, lifespan, weight, and height. A primer panel was designed to target approximately 200bp amplicons surrounding the markers of interest. The panel was designed to target both the Watson and Crick strands of DNA. The sequencing workflow consisted of an initial bisulfite conversion using as little as 10ng of DNA input. Library preparation was then completed using the Thermo Fisher Scientific AgriSeq™ HTS Library Kit. Template prep and sequencing were performed on the Genexus™ Integrated Sequencer, a highly automated system that performs all post-library manipulations without manual intervention. After sequencing, the analysis pipeline provided DNA methylation calls on both Watson and Crick strands at single base resolution and methylated:unmethylated ratios for each targeted CpG marker. The performance of the workflow was evaluated by serially diluting a fully methylated canine control DNA sample into a fully unmethylated sample at the following ratios: 100%, 75%, 50%, 25%, and 0% methylation to demonstrate the accuracy of DNA methylation quantitation across a range of inputs. The workflow was completed in 2 days, end-to-end, and generated highly accurate quantitative results with as little as 10ng of DNA input. The workflow can be adapted for any CpG markers of interest using custom-designed primer pools from the Thermo Fisher Scientific White Glove team highlighting the flexible and customizable nature of the workflow.

SMARTER-database: a tool to explore genomic diversity in small ruminants

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We present the SMARTER-database, a collection of tools and scripts to standardize genomic data and metadata of small ruminant populations. With a focus on reproducibility, our system prioritizes consistency and reliability in data integration. Genotypes are standardized to the same coding convention and to the most recent reference genome assembly version, in order to facilitate seamless comparison of samples from different sources. Data access is granted to users through the SMARTER-backend using R packages or the web interfaces, while genotypes are available over FTPS. Currently, the SMARTER database tracks genomic information on approximately 12,000 sheep and 6,000 goats, with nearly 80% and 60% of samples possessing GPS coordinates of sampling sites of sheep and goats, respectively.

Efficient calculation of linkage analysis based genomic relationship matrices for optimum contribution selection.

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In the pre-genomics era, inbreeding was based on pedigrees and estimated as the probability of Identity-by-Descent (IBD) at neutral loci unlinked to loci under selection. However, with genomic selection for genome-wide dense marker panels unlinked neutral loci hardly exist anymore. With genomic data, we can see what happens at the DNA level and a multitude of genomic measures of inbreeding have been developed. Some inbreeding measures are based on homozygosity, such as runs of homozygosity and excess of homozygosity, causing allele frequency changes towards 0.5 in optimum contribution selection (OCS). Other inbreeding measures are based on genetic drift, which favor the loss of rare alleles over a substantial increase of their frequency in OCS schemes. We argue that measures of inbreeding need to be based on IBD, such as the pedigree relationship matrix *A*, because diversity management based on IBD is neutral with respect to allele frequency changes. Genomic information can be included in the *A* matrix using linkage analysis (Gla). This linkage analysis approach was originally proposed by Fernando and Grossman for a single or few marker loci but is computationally extremely demanding for current dense SNP panels and large national data sets. Moreover, the Fernando and Grossman approach would calculate the Gla relationship matrix for all animals in the pedigree instead of only for genotyped animals such as the genomic relationship matrix (GRM) does. Depending on our needs, the latter may be computationally wasteful. Here we develop a linkage analysis approach to calculate the Gla relationship matrix using 50k SNP genotypes for 10,311 young genotyped cows in a pedigree of 393,129 Norwegian Red Cattle. We compared the estimated relationships to the GRM, the *A* matrix and the inbreeding coefficients to those obtained from Runs of Homozygosity.

Characterisation of grazing dairy cows differing in how they utilise their ingested nitrogen

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Excessive nitrogen loss for agriculture can contribute to pollution. Selecting cows that efficiently use nitrogen (i.e., cows that can utilise nitrogen intake in milk, growth, and pregnancy more effectively) while also excreting (relatively) low quantities of nitrogen can help mitigate this issue; genetic variability in both traits is known to exist. Nitrogen use efficiency and the quantity of nitrogen excreted was available from 491 lactations from 184 grazing dairy cows, totalling 1,249 weekly estimates. Linear mixed models were used to compute animal solutions for both nitrogen utilisation and nitrogen excretion and these were jointly used to classify the cows as sustainable if their nitrogen use efficiency was better than average while also excreting less nitrogen than the average. Conversely, cows were classified as undesirable if their nitrogen use efficiency was worse than the average while excreting more nitrogen than the average. Performance metrics were computed for both classes using linear mixed models. The sustainable cows were 20 kg lighter (SE = 5 kg), ate 53 grams of nitrogen less per day (SE=12 grams of nitrogen/day), and produced 190 grams more protein per day (SE=87 grams/day) than the undesirable cows. Neither milk urea nitrogen nor milk yield differed by class. This study suggests a potential solution for more sustainable grazing dairy milk production.

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Theatre 2

Feed, nitrogen and phosphorus-use efficiency in organic and conventional dairy farming systems in The Netherlands

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Enhancing feed efficiency (FE), nitrogen use efficiency (NUE), and phosphorus use efficiency (PUE) is pivotal for sustainable dairy cattle production. We evaluated FE, NUE, and PUE using Annual Nutrient Cycling Assessment data from 109 organic (OF) and 856 conventional farms (CF) in the Netherlands during 2018-2021, in which CF were selected based on proximity to OF, breed (>80% Holstein-Friesian) and herd size (>50 cows). For each farm and year, NUE and PUE were calculated by comparing farm outputs (milk and meat) to inputs (feed) at herd level (youngstock, dry and lactating cows) and farm outputs (milk, cattle, and sold crops and forages) to inputs (cattle, feed, fertilizer, and N fixation and deposition) at farm level. Data were analysed with a mixed model, including farm type (OF or CF), year (2018 to 2021) and soil type (sand, clay, peat, or mix) and all interactions between farm type, year and soil type as main effects, with farm as repeated effect. Preliminary results show that CF had higher FE (1.16 vs. 1.02 kg FPCM/kg DM) than OF. Furthermore, CF had higher PUE at herd level (35.9% vs. 31.1%), while PUE did not differ between CF and OF at farm level, except for a higher PUE of CF on mixed soil in 2018 (81.4% vs. 70.2%). Lastly, CF showed higher herd NUE (25.1% vs. 23.8%), but lower farm NUE (33.9% vs. 39.5%) than OF across all soil types, except for peat soil where no differences were found. Enhancing knowledge on dairy farming management strategies, dependent on efficiency metrics, may help to decrease excretion of N, P and other pollutants to the environment and decrease the amount of feed used to produce milk.

Carbon footprints result from livestock production in Poland based on NPC tools from CCCFarming project - limitations and perspectives

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The use of accounting tools for estimating greenhouse gas (GHG) and ammonia (NH₃) emissions from dairy farms is of increasing importance for monitoring environmental performance and identifying effective mitigation strategies. In the EU project, Climate Care Cattle Farming Systems ('CCCfarming') three GHG accounting tools were used: ANCA, AgreCalc, CAP2ER. In Poland – one of the project participants – eight selected farms representing a country-wide variety of housing systems, management practices, breeds, feeding, and land were used. One large farm with 1437 cows and the smaller one's with 10 to 100 cows were monitored. Average milk production per cow was 9241 kg (from 5600 in small 10 cows-farm and in ecological to 15 000 in intensive production farms) in all analyzed farms. Compared with the other farms from the other CCCfarming project countries, based on NPC tools, GHG emission intensity in Poland was one of the highest (1620 g CO₂eq/kg FPCM). The average GHG emission intensity in Polish farms was 1.44 kg CO₂eq/kg FPCM whereas in the other project countries from 0.94 to 1.08 kg CO₂eq/kg FPCM. However, it should be emphasized that the implemented NPC tools were adapted to Polish conditions and were not always easy to use, therefore, the results obtained for other countries such as Latvia, or Lithuania may systematically differ. We can however conclude that the emission reduction strategies proposed in the project, e.g. covering slurry tanks or nutritional strategies, can significantly reduce emissions from Polish dairy farms. Project CCCfarming National Centre for Research and Development (SUSAN/II/CCCFARMING/03/2021)

Session 3

Theatre 4

The duration of outdoor stay affects the urinary and faecal nitrogen excretion in dairy cows

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Access to outdoor areas enhances animal welfare in dairy cows. The time and duration of the stay is critical in order to prevent excessive land nitrogen deposition. The objective of this study was to assess nitrogen (N) excretion in urine and faeces of dairy cows during daily outdoor stays of 2 and 4 hours. The experiment was carried out at the Paolino d'Aquileia Institute (Cividale del Friuli, Udine, Italy), which features a free-stall equipped with an automatic milking system. Six (6) Italian Simmental lactating cows were selected based on lactation number (2.0±1.4) and lactation stage (103±35 DIM). Animals were fed a TMR based on grass-silage and alfalfa hay. Using a 3×3 Latin square design with 3 periods of 2 weeks each, the cows were grouped into 3 categories: confinement inside a free-stall all day (CTR), 2-hour daily outdoor access (11:30 a.m. – 1:30 p.m.) (O2), and 4-hour daily outdoor access (O4). The O4 group had two exit times per day: morning from 9:00 to 11:00 a.m. (O4a) and afternoon from 2:00 to 4:00 p.m. (O4b). Urine and faeces, produced during the outdoor stays, were collected, weighed and sampled for N analysis using the Kjeldahl method. Fresh faecal weight, N content in faeces, total N output of faeces and overall N excretion (urine and faeces) were significantly (P<0.01) higher in O4a compared to O4b (3.82 vs 2.17 kg, 2.16 vs 1.69% DM, 13.16 vs. 7.08 g, and 27.97 vs. 16.16 g resp.). O2 group showed intermediate values compared to O4a and O4b. To conclude, due to lower fecal output and reduced concentrations of N in urine and faeces, the optimal time for access to outdoor areas appear to be in the afternoon, specifically between 2:00 and 4:00 p.m. Considering the limits of EU Directive 91/676, the number of cows per hectare could potentially increase from 17 to 22 and 29 respectively for morning, midday and afternoon outdoor outings throughout the year.

The characterization of microbiome and resistome in feces and slurry of dairy cows in different livestock production systems

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The application of manure or slurry to agricultural land is considered an environmentally friendly method of waste disposal (Wen et al., 2019); it allows providing the essential nutrients to the soil and enhancing its quality, improving the yield and quality of crops and pastures, and additionally reducing the use of mineral fertilizers. However, because of the huge antimicrobial use in dairy cows and other livestock (Köchle et al., 2024), these effluents are implicated in the transmission of antimicrobial resistance (AMR) bacteria into aquatic, terrestrial and atmospheric ecosystems (Sura et al., 2015; McNab et al., 2007 and McEachran et al., 2015) which threatens public health on a global scale (WHO, 2014). Here lies the importance of characterizing the microbiome and resistome composition in feces and slurry of dairy cows, comparing three livestock production systems (intensive, conventional grazing; and organic grazing). To achieve these objectives, 18 dairy cattle farms (six farms per production system) were selected in Cantabria, Spain; to whom a questionnaire on general farm characteristics, herd management, effluent management and health management practices was conducted. In addition, 300 feces samples and 90 slurry samples were collected from first-calving Holstein cows and piles of accumulated slurry respectively. All samples were analysed in the CIFA laboratory both for physicochemical and molecular analysis. The metagenomics DNA were quantified and, their quality and integrity were evaluated. The sequencing process was realized using MinIon Mk1C and following SQK-LSK109 protocol of ONT (Oxford, UK). The bioinformatics tool used is the SQM pipeline. The preliminary results showed that the mean read length is 2 731 bp with a median read quality of 14.34 and N50 of 6 935 bp. Metagenomes studied are mostly composed of bacteria (84%), followed by archaea (4%) and viruses (<1%). The most abundant metagenomes in feces are *Methanobrevibacter* sp. YE315, *Bifidobacterium pseudolongum* and *Bifidobacterium angulatum*. In slurry, the most abundant metagenomes are *Nostoc* sp. C057, *Escherichia coli* and *Pseudomonas nitroreducens*. In feces, we found 30 gen groups of AMR and in slurry samples 67.

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Theatre 6

Response of early and late lactation dairy cows to diets differing in levels of human edible ingredients

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Twenty early lactation (EL) cows and 20 late lactation (LL) cows (65 and 220 days in milk) were used in a 2 period (4 weeks duration) changeover design experiment. Four treatments were examined in a 2×2 factorial design comprising two lactation stages (EL and LL) and two concentrate types differing in human edible fraction (HEF) (human edible, HE) and low human edible (IN). HE was predominantly cereal and soyabean meal based, while IN included significant quantities of feeds like wheatfeed, rapeseed meal and distillers dark grains. Concentrates were formulated to be isoenergetic and isonitrogenous. Diets were offered as a PMR (57% grass silage:43% concentrate, DM basis). A standard concentrate (0.5 kg/day) was offered at each milking. Data (from week 4 of each period) were analysed using linear mixed model (REML estimation method) methodology with Cow and Period fitted as random effects and a factorial arrangement of Lactation stage and Concentrate type fitted as fixed effects. Pairwise differences between the levels of the effect were compared using Fisher's Least Significant Difference test. HEF of HE and IN were 0.506 and 0.184 respectively. Concentrate type had no effect on total DM intake (HE, 21.1; IN, 20.6 kg/day, P=0.228) or milk yield (HE, 30.3; IN, 30.3 kg/day, P=0.833). Cows offered HE produced milk with a higher protein content (33.5 g/kg) than cows offered IN (32.5 g/kg) (P<0.001). Irrespective of stage of lactation, calculated edible Feed Conversion Rate (eFCR)(MJ edible output/MJ edible input) was greater for cows on IN than HE (EL-HE, 1.27; EL-IN, 3.31; LL-HE, 1.09, LL-IN, 2.70; P=0.001). In conclusion reducing the HEF in a concentrate had no impact on the DM intake and milk yield of early and late lactation cows but improved eFCR.

Reduced climate change impact of milk achieved with introduction of faba bean in dairy cows' diet

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The impact of substituting typically used imported rapeseed meal (RS) with domestic faba bean (FB) was studied at dietary concentrate levels of 45% and 35% on grass silage (GS) based Finnish dairy cows' diets. The climate change impact (CF) of milk was assessed using life cycle assessment (LCA), using both (a) the IPCC method and (b) the specific methods developed for Finnish conditions, according to Hietala et al. (2021). For each diet the LCA model was adjusted for the diet composition (incl. crop cultivation and nutritional values), milk yield and composition. Milk yield was converted to fat and protein corrected milk (FPCM, IDF 2015). For the FB diets, feed cultivation was modelled including crop rotation and pre-crop effects (i.e., N fertilization, crop yields) as presented by Kuoppala et al. (2024). The number of calvings was assumed to be 3, the slaughter age 1825 d and the slaughter weight 268 kg, for all diets. The RS diets (45RS, 35RS) included 4000 and 4413 kg DM of GS year⁻¹. Corresponding amounts of RS were 650 and 469 kg DM, whereas the amounts of FB substituting RS were 826 and 595 kg DM (45FB, 35FB). All diets were completed with barley and oats in the ratio of 1:1. Increasing the GS and the FB in the diet led to lower FPCM yield. 45RS resulted in 10735 kg FPCM year⁻¹. Substituting FB for RS reduced FPCM by 2% and increasing GS proportion decreased FPCM by 6 to 8%. Lowest CF of FPCM resulted from 45FB with 1.25 (a) / 1.17 kg CO₂eq (b). 45RS and 35FB both resulted 2-3% higher CF and 35RS performed worst with 4-5% higher CF. The introduction of FB led to lower FPCM, yet the environmental performance of feed cultivation was improved with higher rate, overcoming the impaired milk yield.

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Theatre 8

System level impacts of environmental and social economic optimization-expert analyses of different countries

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The European Climate Care Cattle project provided an evaluation of farm systems optimizing socio-economic and environmental outcomes. To achieve this we have utilised research outputs from different work packages and used a workshop of project partners of eight countries to provide expert analysis to evaluate management options for improving the environmental outcomes of European dairy farming. A structured questionnaire was prepared that divided the management of the farm into the following groups: Livestock management, Pasture and soil management, Housing design and manure management, and Energy. The project participants were allocated to each group in accordance with their expertise and region. Also, a limited number of four suitable farm systems were evaluated (Intensive, Extensive, Organic and Visionair) for each region. The various management and farm systems were in a stepwise procedure scored on outcomes related to productivity, the environment and their attractiveness to the region. The systems vary between regions in Europe in order to support locally appropriate farming. This study has highlighted the important role that management interventions have in delivering efficient, productive and low emission dairy production across Europe. Milk produced in European dairy systems already achieve high levels of efficiency when compared with global baseline, however it is also recognised that more needs to be done to reduce current levels of emissions in order to meet ambitious climate change mitigation targets. No single intervention will achieve this, but our study has shown that changes to livestock management, pasture management, housing, energy use and manure management can have beneficial and additive effects on productivity and environmental outcomes. The carbon footprints of real dairy farms across Europe indicate that the variability within regions is often larger than that between regions highlighting the opportunity for management interventions to influence environmental outcomes.

N, P and C efficient use and circularity

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Circularity in nutrient management is an approach aimed at mitigating some of the environmental effects of agriculture, by curtailing nutrient losses through recycling and reutilisation. The global food system contributes 20-30% of anthropogenic greenhouse gases and contributes to fresh and estuarine water body degradation as well as biodiversity loss. Anything that reduces these negative effects is to be commended. Nutrient circularity has focussed specifically on the efficient use of nitrogen (N) and phosphorus (P), because of their recognised importance in food production and their role in ecosystem degradation when improperly managed, and, carbon (C), because of its role in climate change. Agricultural N is sourced from fertiliser, mostly (estimated to be responsible for approximately half of the world's food supply), animal waste, or from fixation of atmospheric N into ammonium by N-fixing bacteria. Phosphorus is generally sourced from finite rock deposits, which are spatially isolated from both food production and consumption locations. Nutrient circularity strategies can enhance nutrient-use efficiency and reduce reliance on non-renewable resources. Adopting sustainable land management practices, like agroforestry and carbon sequestration initiatives, and encouraging changes to consumption patterns to increase natural fibre use in lieu of synthetics (eg wool for carpets, timber for building) may mitigate climate impacts as well as other pollution externalities (eg nano- and microplastic pollution). Areas suitable for food production are often spatially removed from centres of population and, in some cases, sources of nutrient inputs, limiting circularity initiatives. A drive for circularity cannot be allowed to significantly reduce efficient food and fibre production in population sparse areas to encourage food and fibre production in less efficient regions close to centres of human and animal populations. With a growing global population and pressing food and nutrition security needs, agricultural production must be undertaken where it is most nutrient and C efficient. This requires a strong rules-based trade system without barriers to trade.

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New York dairy farms: their whole-farm nitrogen and phosphorus balances and greenhouse gas footprints
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New York (NY) has over 620000 dairy cows on almost 2800 farms producing more than 7 million Mg of milk per year, making it the fifth largest dairy producing state in the United States. For 36 large (>350 cows), commercial NY dairy farms, the Cornell University Whole-Farm Nutrient Mass Balance (NMB) calculator was used to calculate nitrogen (N) and phosphorus (P) NMBs, and Cool Farm Tool was used to estimate greenhouse gas emission intensity (GHGei) for the 2022 calendar year. Farm size ranged from 350 to 6000 head of predominantly Holstein cows with animal densities between 1.76 and 4.85 animal units/ha, average fat and protein corrected milk (FPCM) yield of 12.7 Mg FPCM/cow/year and an average of 63% homegrown feed. Total FPCM production was 870000 Mg, representing approximately 12% of total NY milk production in 2022. Balances for N ranged from 9 to 287 kg N/ha (weighted average = 151 kg/ha) and 0.4 to 14.6 kg N/Mg FPCM (weighted average = 7.7 kg N/Mg FPCM). Balances for P ranged from -18 to 30 kg P/ha (weighted average = 12 kg P/ha) and -0.9 to 1.6 kg P/Mg FPCM (weighted average = 0.6 kg P/Mg FPCM). The GHGei ranged from 0.63 to 1.06 kg carbon dioxide equivalents (CO₂e)/kg FPCM (weighted GHGei = 0.81 kg CO₂e/kg FPCM). These results show good management of both N and P for these NY dairy farms, although opportunities remain for improvement. Both N and P weighted balances per Mg FPCM meet the feasible limit set for NY dairy (8.8 kg N and 1.1 kg P/kg FPCM), indicating high FPCM production efficiency. Weighted balances per ha meet the feasible limit set for P (13 kg P/ha), but exceed the feasible limit for N (118 kg N/ha). For GHGei, current performance reflects the high proportion of good quality homegrown feed, careful nutrient management and use of manure treatment systems such as covered liquid slurry storages and anaerobic digesters on these dairy farms. These results represent a considerable proportion of NY's 2022 FPCM production. However greater participation by smaller farms with less intensive milk production is necessary to draw conclusions on NY's dairy industry as a whole.

Potential use of drones and ICT in livestock farming to deal with environmental sustainability

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The need to improve environmental sustainability and reduce nutrient losses in livestock farming is driving the adoption of ICT-based tools in animal production. The utilization of advanced sensor networks, data analytics and precision farming techniques holds the promise of enabling real-time, granular monitoring of both animal and environmental performances. Integrating ICT with intelligent decision support systems would, therefore, optimize resource utilization while minimizing the environmental impact of farming activities. The objective of this study was to offer insights into the potential use of drone-based techniques and ICT in the environmental assessment of farms. This was based on the findings of a literature analysis and the results of a research project focused on developing a drone-based system for identifying on-farm gas and dust emissions. The system incorporated prototype multi-sensor systems for measuring CH₄, CO₂, NH₃, PM_{2.5}, and PM₁₀ at ground level and during flight. A real-time system for collecting, storing, and visualizing data was developed. Additionally, modelling tools for quantifying and mapping emissions were selected and tested. The results of this study showed that, despite the existence of theoretical models and sensor technologies enabling the rapid measurement, estimation, and mapping of gas and dust emissions at the farm level, the system requires optimization to ensure reliable results. The development of effective ICT tools relies significantly on lightweight and highly accurate sensors. Additionally, it involves testing and selecting suitable methods for quantifying emissions in environments marked by multiple sources of emissions and turbulence.

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Theatre 12

Reducing slurry temperature to lower NH₃, N₂O and CH₄ emissions from fattening housing

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The aim of this study was to determine the influence of low slurry temperatures on gaseous emissions and to compare two manure cooling systems. The study was conducted on three fattening rooms each housing 54 pigs between 30 and 110 kg. For the three rooms, the design was strictly the same in term of type of floor (fully slatted floor), feeding strategy (biphase – 16 and 15% of crude protein) and ventilation management (diffuse ceiling, under-floor extraction, identical ventilation set). One room (MCSb) was equipped with a manure cooling system (MCS) with pipes fixed to the bottom of the pit. In the second one (MCSf), the cooling pipes were fixed on a raft floating on the surface of the slurry. The last room was the reference (Ref) with no cooling system. Pigs were individually weighted. Feed and water consumptions were daily recorded. Ambient and slurry temperature (three different heights in the pit) were recorded each hour. Slurry volume per room was frequently measured and sampled for analysis. NH₃, N₂O, CH₄ and CO₂ concentrations were semi-continuously measured with a photoacoustic analyser (Innova 1512, Lumasens Technologies Inc.). Carcass performance were also individually recorded (meat rate, fat and muscle thickness). No significant effect was observed on pig performance (average daily gain, feed conversion ratio) and carcasses. Concerning gaseous emissions, NH₃ was reduced by 20% in the MCSb room and less than 10% in the MCSf room. No effect was observed on N₂O. The reduction of CH₄ was around 70% in the MCSb room and less than 30% in the MCSf room. Reducing slurry temperature can be efficient on NH₃ and CH₄ emissions but the efficiency of the MCS with pipes fixed on the bottom was greater than the floating system.

Response of grazing dairy cow production to different grass swards under varying nitrogen fertiliser rates
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A grazing study was undertaken in 2022 and 2023 to determine if white clover inclusion in a grass sward could decrease nitrogen (N) fertiliser rates while maintaining a similar production to a grass-only sward under a high N fertiliser rate. The grazing study compared two sward types under two rates of N fertiliser. The four treatments were a grass-only sward receiving 150 kg N/ha (GO-150) or 225 kg N/ha (GO-225) and a grass-white clover sward receiving 150 kg N/ha (GC-150) or 75 kg N/ha (GC-75). Lactating dairy cows were randomised and blocked into treatments by breed, parity, and calving date (n=112). Each treatment group was stocked at 2.57 cows/ha and rotationally grazed 20 paddocks. Grass measurements were taken before and after grazing. Milk yields were recorded daily and milk composition was measured once per week. Statistical analysis was undertaken using mixed models with fixed effects including treatment, paddock, year, and block for grazing data and treatment, breed, parity, and year for milk data. The GO-225 and GC-150 had a similar (P=0.71) total herbage dry matter production. Cows grazing the GC-150 treatment had greater total milk solids (fat + protein) yield (P < 0.05; -20 kg/cow) than cows grazing the GO-225 treatment and tended to have greater total milk solids yield (P = 0.058; -19 kg/cow) compared to cows grazing the GO-150 treatment. The results show a reduction of 75 kg N/ha and the inclusion of white clover can achieve similar herbage and milk production as a grass-only sward with higher N inputs.

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White clover's positive impact in perennial ryegrass swards with reduced nitrogen inputs on milk production and nitrogen use efficiency

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To reduce cost inputs and environmental impacts of inorganic nitrogen (N) use, there is renewed interest in the incorporation of legumes, white clover in particular in grazing systems. The objective of this study was to reduce chemical N fertiliser input to temperate grass-based milk production systems by incorporating white clover into grassland swards and in turn increase farm gate N use efficiency (NUE). Three grazing treatments were used for this study; grass-only swards receiving 200 kg N/ha (Gr200) and grass-white clover swards receiving 100 (C1100) or 150 kg N/ha (C1150) annually. Twenty cows were assigned to each treatment and swards were rotationally grazed at a stocking rate of 2.5 cows/ha. In the first year of the study, cows grazing the grass-white clover treatments had greater production in terms of milk yield (+257 kg) and milk solids yield (+35 kg) and NUE (+23%) compared with Gr200 treatment. The grass-white clover swards produced 5,858 kg milk and 515 kg milk solids (kg fat + protein) per cow. There was no treatment effect on herbage production, 14.8, 14.9 and 14.9 tonnes DM/ha for Gr200, C1150 and C1100 systems respectively. This significant increase in milk production suggests the inclusion of white clover in grazing systems can be effectively used to increase milk production and reduce N fertiliser inputs and their environmental impacts.

Analysis of nitrogen fertilizer efficiency in rotational grazing: Impact on the quality and productivity of *Lolium multiflorum* Lam. Forage

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Pastures play a crucial role in sustaining dairy production, particularly in temperate climates where animals can graze year-round. This study aimed to assess *Lolium multiflorum* Lam pasture quality, productivity, and production costs. Conducted on a farm in São Miguel Island, it compared conventional nitrogen fertilizer with controlled-release nitrogen fertilizer at two levels (320 and 160 kg N/ha), alongside a control group. An annual assessment aimed to discern potential agronomic and economic advantages of controlled-release fertilizer. Using a 5×5 Latin square design, the study had five treatments: high fertilization with conventional fertilizer, high with controlled-release, intermediate with conventional, intermediate with controlled-release, and a control. Results showed fertilizer type did not significantly impact forage productivity or quality, but nitrogen levels significantly affected them. Higher nitrogen doses increased dry matter and crude protein. Fiber characteristics remained relatively stable across most cuts, although higher nitrogen application led to elevated levels of NDF and ADF without affecting ADL. Furthermore, nitrogen use efficiency consistently exceeded 70%, regardless of fertilizer type or quantity. Regarding cost per kilogram of dry matter and crude protein, control groups and conventional fertilizers were more economically efficient than controlled-release. This highlights fertilization's importance in dairy production and the need for economically viable choices. While conventional fertilizers proved cost-effective, controlled-release options lacked comparable economic benefits.

Session 3

Poster 16

Variation of in vitro utilisable crude protein in different red clover accessions

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Red clover silage is widely used for dairy cows, especially in organic farming due to its higher CP content and lower fertilizer requirement compared to most grasses. However, high polyphenol oxidase (PPO) mediated protein complexing, which protects proteins from degradation during ensiling could also affect protein degradation in the rumen. Twenty-three diploid and 22 tetraploid red clover accessions, including two check cultivars of each, from genetic selection experiment at Lännäs, Sweden, were screened for the in vitro utilisable CP value estimated for 16 h rumen retention time (uCP16). Wilted red clover was chopped and ensiled in 1 L vacuum bags with acid-based silage additive added at a rate of 6 ml/kg. Mean DM content of silages was 240 g/kg (SD=32). Tetraploid accessions had higher ($P<0.001$) NDF concentration than diploids, 333 vs 295 g/kg DM, respectively. Mean concentration of CP and uCP16 in diploid accessions were 199 (SD = 11.5) and 168 (SD=7.3) g/kg DM, and in tetraploids 183 (SD=9.5) and 169 (SD=8.4) g/kg DM, respectively. No differences in CP and uCP16 between experimental and check cultivars was measured. uCP16 was highly correlated ($R=0.81$) with CP concentration, however four silages had 2.1-3.9% higher (P values between 0.01-0.06) uCP16 concentration despite no differences in CP concentration. These results imply that protein value varies between red clover accessions, and further results are warranted to evaluate the effect of PPO mediated quinone complexing on protein value.

Success with better calves starts well before birth

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Breeding tools to help breed better calves have significantly enhanced over recent years. It may seem obvious, but breeding replacement calves only from the best females, and mating them to the best bulls will significantly improve the quality of calves born on farm, but can equally create issues if not adopted correctly. Dead calves have an immediate negative impact on the carbon balance of the herd whereas live but poor calves also impact the herd over time. There are many genetic tools available now for producers to use, and this paper will highlight and explain the various genetic indexes available to breed for success and address the additional breeding tools that should be used to maximise opportunities such as genomics and sexed semen. With an industry under pressure to improve efficiencies as well as welfare and its green credentials, the breeding decisions made today will have to be right to ensure success for the herd of the future, since those calves will be in the herd for the next 6.5 years on average, and their genetics will persist in subsequent generations. Latest statistics from the UK industry, which are likely mirrored elsewhere, indicate that dairy farmers have been quick at responding to new signals and information. Use of genomic young bulls now stands at about 70% of all dairy insemination, and female genomic testing has risen to about 20% of all calves born in 2023. The use of sexed semen to further accelerate gains is at well over 75% of dairy breedings, resulting in over 50% of insemination in the dairy herd taken up by beef sires. Genetic trends indicate good progress across a range of traits and will help us with breeding a healthy crop of calves. But are there areas we can do even more in the future? This paper will propose some.

Session 4

Theatre 2

Adequate milk feeding of dairy calves

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Calf growth and development depend on adequate milk intake. Lifting traditional milk restriction in calves has been causally associated with beneficial effects on growth, health and future performance. In this critical early life period, nutrition triggers plasticity of development, ultimately leading to an altered adult phenotype. Although rumen function accelerates with limited milk intake (approximately 6 L milk/day), calf development does not accelerate. Adequate, i.e., ad libitum milk feeding, means an intake of up to 20% of bodyweight as milk per day. Holstein calves possess the ability to digest 12-15 litre of milk daily. Growth performance and organ maturation are improved when calves are allowed to drink greater amounts of milk. Accelerated growth development in calves fed adequately with milk was confirmed by stimulation of the somatotrophic axis, indicating the initiation and continuation of anabolic processes. Adequate milk feeding has an immediate positive effect on feeding behaviour in calves. Calves fed only 6 litres of milk per day showed permanent hunger. Calves with adequately high milk intake may be more resistant to infectious diseases, leading to a reduction in calf loss during the neonatal and preweaning periods. Although elevated milk feeding delays rumen development by reducing solid feed intake, rumen growth is not impaired, and forestomach digestion matures better when more time is spent on the weaning process, and weaning should start later in life. This weaning protocol corresponds to the natural situation in beef calves and avoids hunger and discomfort. Improving calf feeding management will contribute to successful calf rearing by meeting calf demands and raising robust and resilient animals.

Growth performance of calves fed milk replacer or dams milk during the first week of life

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Milk contains many different bioactive compounds especially after parturition which have been shown to play an essential role in calf development, and especially postnatal gut development. The hypothesis was that during the first week of life a switch from the more nutrient dense milk to a commercial milk replacer (MR) results in less growth performance. To test the hypothesis twenty calves of the brown swiss breed were separated from their dams directly after birth and housed individually for the first week of life. They were fed twice daily with nipple buckets with unlimited milk amounts from their dam. Upon the 7th feeding ten animals were randomly chosen to transition to MR feeding with 150 g of MR per liter (13% dry matter). The other ten animals continued to receive their dams milk. The animals were weighed at the beginning of the experiment, before the 7th feeding and at the end of the experiment. Performance parameters were calculated for the whole week and statistically analysed for differences in group means using a t-test. All animals consumed high amounts of milk and there were no differences between the treatments ($p = 0.62$). The mean daily growth rate of > 1 kg for both treatment groups showed that all animals achieved satisfactory growth. Calves switched to MR feeding showed less average daily gain than the milk fed ($p = 0.004$). Also, when expressed relative to birthweight MR fed calves showed less relative body weight gain throughout the week than milk fed calves ($p < 0.001$). During the first week of life, calves fed their dams colostrum with subsequent milk feeding showed superior weight gain compared to calves fed colostrum and consecutive MR feeding.

Session 4

Theatre 4

Evaluating early life feeding strategies on the development and health of pre-weaned surplus calves

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This study examined early-life interventions and their impact on health and development in Holstein-Friesian surplus calves. In the first 12h after birth, 80 bull calves (43.1±3.56 kg BW) were fed either 2L (100g of IgG, F) or 6L (300g of IgG, S) of colostrum replacer (CR). Upon arrival at the facility, serum IgG was measured and calves were blocked based on CR protocol, arrival date, age, and BW. Within each block, calves were randomly assigned to one of two milk replacer (MR) feeding: Moderate (Mod;~8L) or Low (Low;~4L). In total, there were 4 treatments resulting from the 2×2 factorial design: F-Mod, F-Low, S-Mod, S-Low (n=20/trt). Calves were individually housed and received their MR allowance twice daily in teat buckets. Calves had ad libitum access to straw and water during the entire study, and on d22, a calf pelleted starter was offered. On d42, 10 calves/trt were euthanized. Intakes, and fecal score were measured daily, and BW was measured weekly. Data were analysed by PROC MIXED in SAS accounting for the fixed effects CR, MR, time, and their interaction, as well as the random effect of block. Time entered the model as a repeated statement. Serum IgG was greater in S calves (S=19.6 vs. F=9.4 g/L). Overall, Mod calves had higher BW despite CR treatment ($P<0.01$). Fecal consistency was impacted by CR ($P<0.01$) and MR ($P<0.01$), with S and Low calves having better consistency. Moreover, F-Low calves had the highest number of treatments for diarrhea ($P=0.04$), while F calves had the most treatments for fever ($P<0.01$). At d42, calves receiving Mod levels were on average 13.2 kg heavier than Low calves ($P<0.01$). Increased fresh tissue weights were observed in the small and large intestine of Mod calves ($P<0.05$), regardless of CR treatment. These findings highlight the importance of considering both colostrum and MR feeding levels in calf nutrition to optimize growth, development, and health during early life.

Observations on neonatal calves' urine excretion during the first week of life

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Proteinuria is known to occur shortly after birth in calves, since the kidneys are partly immature. Therefore, low molecular weight proteins can be found in the postnatal urine. This study aimed to investigate urine excretion patterns in neonatal calves with regard to a change in feedstuff. Twenty brown swiss calves were separated from their dams at birth and housed individually for the first week of life. All were fed their dams colostrum for 6 feedings following either transition milk (n=10) provided twice daily with unlimited volume or calves were transitioned to milk replacer (MR) (n=10) from the 7th to the 14th feeding onward. Urine was quantitatively collected using canisters. The weight of milk ingested and urine excreted was then divided and the ratio statistically analysed using a two factorial ANOVA with time and treatment as fixed factors. The urine excretion of newborn calves exhibited a polynomial trend, gradually tapering off towards an asymptote where the volume of urine excreted approached approximately half the amount of milk ingested by the end of the week. Directly after birth, the ratio of ingested milk and excreted urine is very broad with only small amounts of urine excreted compared to the amount of milk ingested. From the 7th feeding onward a significant effect of treatment on the milk-urine ratio was found (p=0.04) where the ratio was more narrow for MR fed calves. Neonatal calves show a high fluid disappearance rate immediately after birth, indicating a dehydration. This state rapidly disappears within the first days and towards the end of the first week, nears a constant excretion of urine of about half the amount of milk ingested.

Session 4

Theatre 6

Predicting pre-weaned dairy calf weight from morphometric measurements

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Regular weighing of dairy calves is an essential management practice for dairy farmers that contributes to health, welfare, and overall productivity within the dairy operation. Weighing calves may not be feasible due to logistical constraints. Thus, alternative methods that can still help monitor calf growth are valuable. This study aimed to develop a body weight (BW) predictive model for pre-weaned calves using morphometric measurements (MS). From 4 dairy farms, 209 Holstein-Friesian calves aged from 1 to 90 days (188 females; 53 males) were included in this study. Calves were weighed with a digital scale, and heart girth (HG), abdominal girth (AG), withers height (WH), hip height (HH), and body length (BL) were measured. Regression models were run with all MS, considering sex and farm. The predicted weight was compared with BW using Bland-Altman plots and ICC. To increase applicability, a web application (WAp) was developed to allow farmers to easily apply the developed equations. BW ranged from 18.5 to 129.5 kg (54.6±15.6 kg). All MS were correlated with BW and showed a linear or quadratic relationship with BW. HG had the highest correlation with BW (r=0.97). Sex of the calf and farm significantly influenced the relationship between MS and BW. A quadratic model with HG and HH had the best fit (R²=0.97). An overall quadratic model with HG alone showed very similar results (R²=0.96) and was better in agreement measurements than models separated by sex. This model had a better relationship between predictive ability and feasibility and was included in the WAp.

Ultrafiltration enhances the passive immunity properties of low-quality colostrum

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An insufficient availability of high-quality colostrum contributes to a high prevalence of failure in passive transfer of IgG in calves, having negative impact on calf health and long-term productivity. This study aimed to investigate whether ultrafiltration using vibration-shear technology, enhances the passive immunity properties of low-quality colostrum. A total of 25 colostrum samples were divided into five groups based on their initial IgG concentration: IgG-Grp I: 10.0-19.9, IgG-Grp II: 20.0-29.9, IgG-Grp III: 30.0-39.9, IgG-Grp IV: 40.0-49.9, and IgG-Grp V: 50.0-59.9 g/L of IgG. Colostrum was ultrafiltrated by a 30 kDa membrane, using a Sani Membrane VibroETM-LE system, involving a vibration device inducing turbulence at the membrane surface. Two treatments, non-processed colostrum (NPC) and ultrafiltrated colostrum (UFC) and the five IgG groups were included in the experiment. The effect of ultrafiltration on the composition of colostrum was analyzed using a linear mixed model, considering colostrum processing (NPC vs. UFC), IgG-Grp and their interaction as fixed effects, and colostrum batch included as a random effect. An average volumetric concentration factor of 1.77 was achieved. A significant interaction was observed between colostrum treatments and IgG grp for Brix%, IgG, protein, and fat concentrations, with the largest increase in IgG-Grp I (100, 138, 111, and 60%, respectively). In contrast lactose decreased. Colostrum with IgG > 20 g/L seemed most suitable to reach high-quality status (i.e., IgG > 50 g/L). This result allows for feeding of ultrafiltrated low-quality colostrum to newborn calves, ensuring successful passive immunity transfer.

Impact of growth rate of dairy heifers on long term performance: a quantile regression analysis

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Heifers symbolize the future of dairy farms. Their development during the rearing period can be influenced by multiple factors. Although literature suggests that development or growth rate may impact production performance during later life, on farm growth monitoring is mostly incomplete (limited number of weighing times). This challenges the evaluation of the relation between growth and performance for on field data. Therefore, we used a quantile regression analysis to evaluate the impact of growth rate on 1st lactation performance. First, on 30 Flemish dairy farms all heifers were weighted and measured quarterly for 2 years. Second, fertility and milk performance were collected by the dairy herd improvement data. Third, a 10% quantile was designated by the nearest 10% regression line for each measurement of a heifer. During further analysis, the mean quantile of a heifer, representing the growth rate, was correlated with her fertility and milk production data using in total 13.500 on farm follow-up measurements. The 25% greatest growers (>P75) calved 1.1 month earlier in comparison with 25% weakest growers (<P25) (P<0.001). The >P75 heifers produced 960 kg more milk (305D production) during 1st lactation than <P25 (P<0.001). Fat and protein content were 0.08% (P=0.004) and 0.03% (P=0.003) lower in >P75 heifers, respectively. However, due to the higher milk quantity, the impact on content was economically negligible. In conclusion, heifers with a higher growth rate have an earlier age at first calving and a higher production, which is possible related to a better udder development. Quantile regression analysis (here used for the first time) is a very comprehensive way to evaluate the impact of growth rate on production performance of dairy heifers.

Tube ventilation systems for optimizing stable climate in calf barns

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Modern housing systems have to fulfill the individual needs of calves. These systems are inextricably linked to year-round, safe and optimised ventilation of the barns. Calves and young cattle in particular have limited thermoregulatory behaviour that need to be taken into account for a better health status. In this context the year-round suitability of two automatic controlled vet.smart.tubes with FN035 and FN045-EC fans (Ziehl-Abegg) were examined in a barn for 45 calves and young stock (FV x RF). The conditioning showed significant effects in SPSS between outside temperature ($M=27.51$ °C) and air temperature in the surrounding of the calves ($M=21.43$ °C) from July to September 2022 in a daily, 10 minute measuring interval from 12 to 6 pm ($p<.001$, $n=1\ 627$) with a middle flow rate inside the tube of 2.03 m/sec. and 0.29 m/sec. in the calf environment to avoid heat stress. The calves vitality remained consistently high due to a perfect management (removal of manure and renewing deep straw bedding once a week; watercleaning of all boxes every third week), a high amount of whole milk (start 14 l/d) and optimization by tube ventilation. Contrary expectations the animals coped very well in the months January to March 2023 despite low temperatures in daily 24h measurements (minimum=-13.10 °C in the supply air, $M=0.91$ °C and a minimum of -7.70 °C in the calf environment, $M=3.18$ °C, $n=9\ 073$) by avoiding draught and very low amounts of carbon dioxide and ammonia in the barn. The positive effect of the constant air circulation ($M=0.59$ m/sec. inside the tube and 0.23 m/sec. in the barn) was seen in an optimized relative humidity ($M=61.95$ % around calves to 72.27 % outside, $p<.001$) for a hygienic housing environment. During the entire case study there was only one calf showing illness (lung disease). In cooperation with the Austrian Centre for Animal Husbandry and Animal Welfare the tested tube ventilation system can be recommended for calves and young stock under clearly specified installation and operating conditions.

Session 4

Theatre 10

Relationship between growth parameters of heifers at various stages of rearing and milk production in first lactation

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The aim of the study was to determine whether body weight (BW) and average daily gain (ADG) at various stages of rearing affect first lactation milk yield (1MY). Data on BW at birth (BW1), weaning (BW2), 6 (BW3) and 12 months of age (BW4) and ADG between those BW control points (ADG1, ADG2 and ADG3, respectively) of replacement heifers were received from one dairy operator (Ośrodek Hodowli Zarodowej Osiećiny Sp. z o. o.; Osiećiny, Kuyavian-Pomeranian Voivodeship, Poland). Data on pedigree and 305-d milk in 1MY for each animal was obtained from FEDINFO Polish National Milk Recording System. Prior to analysis, genetic effect of each animal was removed from the data set. BW data and ADG in each phase of rearing were regressed against 305 day 1MY, in order to test for linear and quadratic relationship between those variables. 1MY tended ($P = 0.08$) to increase quadratically with increasing BW1, increased ($P < 0.01$) quadratically with increasing BW2 and increased ($P < 0.01$) linearly with increasing BW3. 1MY increased ($P < 0.01$) quadratically with increased ADG1 with no further positive impact on observed when ADG was greater than 1 kg/day, increased ($P < 0.01$) linearly with increased ADG2, and decreased ($P < 0.01$) quadratically with increasing ADG3 with the lowest 1MY observed when ADG was ~ 1.2 kg/day. The relationship between BW and ADG of heifers and 1MY is not linear at all stages of rearing.

Selection for the price of young calves in Italian Simmental breed

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Italian Simmental (IS), Pezzata Rossa Italiana, is a dual-purpose cattle breed, that counts about 90.000 cows registered in its official Herdbook. Selection for beef purpose is based on data collected on young bulls in performance test and the muscling score that is registered by technicians on primiparous milking cows. The selling of calves is a source of revenue for farmers. A tool to select animals for the commercial quality of the calves is not currently available. Kovieh Cooperative is an important wholesale cattle organization operating in Bolzano province (Trentino–Südtirol region, northeast Italy). Kovieh collects purebred and crossbred calves from several dairy herds and sells them individually during public auctions every week. Body weight at sale (BW), price per kg, and the economic value of the calves were collected and registered. The aim of this study was to estimate genetic parameters for the price per kg, which can be considered as a measure of the quality of IS purebred calves. After data editing, 51,825 records of calves' price sold between 10 to 40 days of age were available. Age at sale, BW of calves, and price per kg were 25.9±6.9 days, 70±9.0 kg, and 5±0.69 €/kg, respectively. The model accounted for the auction date, seller, age at calving of the mother, sex, and age at sale (covariate) as fixed, and for direct and maternal as random effects. Heritability for direct genetic effects was 0.21±0.01. The maternal component explained about 2% and there was no correlation between the two genetic effects. Considering these genetic parameters, a genomic evaluation for the quality of IS based on data from auctions could be feasible.

Session 4

Theatre 12

Effects of undernutrition and hydroxytyrosol supplementation during last third of pregnancy on the immune status of suckler cows and their calves

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We aimed to determine the effect of hydroxytyrosol (HT, antioxidant from olive leaves) on immune status of undernourished pregnant cows and their offspring. From 28th gestation week (w) to calving (w40), 136 cows were allocated to four groups (feeding (100 vs 60% requirements) x HT (Control vs HT, for 0 and 178 mg HT/kg uni-feed). Calves were fed dam colostrum and milk. Colostrum samples were collected in Period 1 (0-12h postpartum (pp)) and 2 (12-24h pp). Blood samples were collected in dams (w37 gestation and Period 1) and calves (48h pp and 1 month pp). Immunoglobulin concentrations were determined (Bovine IgG and IgM ELISA, Bethyl, Montgomery, USA). Data were analysed with a mixed linear model with feeding, HT, gestation week and dam age as fixed effects, and dam as random effect. IgM and IgG in cows (plasma and colostrum) and offspring (plasma) were not influenced by feeding level. HT inclusion (HT vs Control) increased colostrum Ig in Period 2 (2.1 vs 1.5 ng/mL for IgM; 73.6 vs 56.4 ng/mL for IgG; P<0.05). Dam age (< 10 vs. >10 year old) affected dam Ig (2.2 vs 1.8 ng/mL IgM, P<0.05; 20.9 vs 27.2 IgG; P<0.001) and calf IgM (0.5 vs 0.4 ng/mL, P<0.05). Ig decreased throughout the study (P<0.001): from w37 to calving in dam plasma IgG; Period 1 to 2 in colostrum (IgM, IgG); 48h to 1 month pp (IgM, IgG) in calves. In short, HT inclusion increased IgM and IgG in colostrum in Period 2, being a strategy to avoid critical Ig decrease in colostrum during the first hours after calving and ensure optimal transfer of passive immunity from dam to calf. Funded by PID2020-113617RR-C21 FETALNUT. Research group A25-23R.

Undernutrition and hydroxytyrosol supplementation during the final third of gestation modulate immune response and redox balance in newborn beef calves

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Hydroxytyrosol (HT) is a polyphenol present in olive leaves that can enhance antioxidant capacity and may mitigate the negative impact of prepartum maternal nutrient restriction in beef cattle. This study assessed the effects of maternal undernutrition (100% vs. 60% of nutritional requirements) and HT supplementation during the last third of pregnancy (Control vs. HT, 178±36 mg/kg of feed) on the whole blood gene expression of key markers related to redox balance and immune response in their offspring during the first week of life. Blood samples were collected from 43 calves in Tempus blood RNA tubes (Applied Biosystems) at week 1 after birth. Gene expression of SOD1, CAT, GPX1, NRF2, NFKB, TLR4, and TNF α was analysed using qPCR. A general linear model including fixed effects (nutrition and HT supplementation) and their interactions was used to compare gene expression with JMP software. A higher CAT and GPX1 expression were observed in calves born from HT-cows compared to those born to Control-cows ($P < 0.05$). Meanwhile, higher TNF α expression was observed in the calves born to 60%-cows compared to 100%-cows ($P < 0.05$). Our results suggest that maternal HT supplementation enhances antioxidant defences responsible for the control of intracellular H₂O₂ levels of the newborn offspring. In addition, maternal undernutrition increased proinflammatory cytokines and therefore might contribute to immune disturbances postnatally. Overall, undernutrition and HT supplementation modulated immune response and redox balance in newborn beef calves. Funded by PID2020-113617RR-C21/22 FETALNUT.

Interplay between pre-partum nutrition and hydroxytyrosol supplementation on calf behavior

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Pre-partum maternal diet restriction and polyphenol supplementation may have carry-over effects on neonatal calf behavior. The role of dietary restriction (100 vs. 60% of the cow nutrient requirements during the last 3 months pre-partum) and hydroxytyrosol (HT) addition (0 vs. 178±36 mg/kg of feed) on beef calves' behavior was evaluated. Twenty-nine (± 7)-day old calves ($n=48$) from four pre-partum maternal dietary treatments (100%-CTR, 100%-HT, 60%-CTR, 60%-HT) were housed in individual pens, with visual and olfactory contact, between suckling periods (08:00-14:00h). Each pen was provided with a warm milk replacer and water bucket with nipple. Observations were performed in three 1-h sessions (9:00-10:00; 10:30-11:30; 12:00-13:00). Calf posture and behavior was recorded by scan sampling every 2 min. The data were analyzed with non-parametric Wilcoxon tests; means were separated with a t-Student test. Calves from 60%-HT cows remained lying for shorter than calves from 100%-HT (33.0 vs. 42.4±2.47 min/h, $p < 0.05$). This led to lower inactive time in 60%-HT than in 60%-CTR (29.7 vs. 37.6±2.41 min/h, $p < 0.05$), more time spent on oral or olfactory manipulation of the pen fixtures (8.2 vs. 5.1±0.97 min/h, $p < 0.05$) and higher number of vocalizations per session (1.5 vs. 0.3±0.37, $p < 0.05$). The 60%-HT calves explored for longer the milk and water bucket than the 100%-HT calves (2.1 vs. 0.6±0.41 min/h, $p < 0.05$), but calves did not perform any nutritive activity therein. In conclusion, maternal nutrient restriction, together with HT supplementation, enhanced calf response to environmental stimuli. Funded by PID2020-113617RR-C21.

Effects of polystyrene nanoparticles on bovine oocyte in vitro maturation

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Oocyte in vitro maturation (IVM) is a critical process in assisted reproductive technologies and is highly susceptible to external environmental factors. The aim of this study was to investigate the effects of polystyrene nanoparticles (PS-NPs) on bovine oocyte nuclear and cytoplasmic maturation. Cattle immature oocytes, obtained from abattoir ovaries, were divided into 5 groups, and matured for 22 h in presence of 0-5-50-100-200 µg/ml of PS-NPs (size 100 nm). Exp. 1: after IVM, oocytes were stained with Hoechst 33342 to evaluate nuclear maturation rate. Exp. 2: after IVM and in vitro fertilization (IVF), oocytes were stained with Hoechst 33342 to assess cytoplasmic maturation. Exp. 3: after IVM, oocytes were stained with CellTracker Blue or H2DCFDA to determine intracellular levels of glutathione (GSH) and reactive oxygen species (ROS) respectively. Data were compared using Chi Square test or GLM and Dunnett post-hoc (IBM SPSS Statistics, significance at P<0.05). A higher rate of matured oocytes was observed in PS0 (62.6%), PS5 (56.9%), and PS50 (59.4%) than in PS100 (31.7%), and PS200 (35.0%), and the degeneration rate was increased in PS200 (20.0%) than in PS0 (9.3%), PS5 (8.3%), and PS50 (6.6%), while PS100 (13.9%) showed intermediate values. No differences were observed in IVF parameters and GSH or ROS levels. These results show that 100 nm PS-NPs at 100 and 200 µg/ml can inhibit bovine oocyte nuclear maturation and induce oocyte degeneration. Although there is still lack of information about the size and concentration of PS-NPs present in mammal tissues and organs, their accumulation along the food chain and ease of penetration are well documented. In conclusion, PS-NPs might be considered a threat for female fertility.

Session 4

Poster 16

Effects of undernutrition and hydroxytyrosol during last third of pregnancy on newborn vitality and cortisol levels of cow-calf pair

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Undernutrition in late pregnancy is a common scenario in extensive systems. Hydroxytyrosol (HT) is a polyphenol present in olive leaves with antioxidant properties. We aimed to determine effects of undernutrition and HT supplementation on newborn vitality and cortisol levels of pregnant Parda de Montaña (Pa) and Pirenaica (Pi) cows and their offspring. Cows (n=109) were allocated to 4 groups (feeding level (100 vs 60% requirement) x HT (0 vs 178 mg HT/kg unifeed)) from the 28th week (w) of gestation to calving (w40). A calf vitality test was performed at birth. Cortisol was determined in blood samples in dams (gestation w37; lactation w1) and calves (w1; w4 of age). A chi-square test for vitality (feeding, HT, breed, type of calving, calf sex, dam age) and a mixed linear model for cortisol (feeding, HT, breed, time as fixed effects, animal as random effect) were performed. Dystocia reduced calf rate with optimal attitude (68 vs 99%; P<0.001), strong finger suckling reflex (73 vs 91%; P<0.05), optimal mucous color (73 vs 94%; P<0.01) and non-protrusive tongue (70 vs 98%; P<0.01). Undernutrition increased cortisol levels in calves (27.6 vs 19.2 nmol/L; P<0.01) but not in dams. Cortisol was higher at calving in dams (12.4 vs 17.5 nmol/L, for w28 and w40) and calves (31.9 vs 14.9 nmol/L; P<0.001). Cortisol values were greater in Pi dams (16.8 vs 13.1 nmol/L; P<0.05) and calves (27.0 vs 19.8 nmol/L; P<0.01). In short, undernutrition in last third of pregnancy increased cortisol levels only in calves. Calves from dystocic parturitions showed the weakest vitality. Funded by PID2020-113617RR-C21 FETALNUT. Research group A25-23R.

Serum and salivary immunoglobulins associations in newborn calves

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This study aimed to evaluate the association between serum and salivary immunoglobulins (Igs) in newborn calves (n=20). Saliva and blood samples were collected at birth (D0; before colostrum intake) and at day 1 (D1), 2 (D2), and 7 (D7) after birth to determine total protein (TP), IgG, IgA, and IgM. Body weight (BW) was recorded at birth. Colostrum samples were collected for chemical composition and Igs analysis. Salivary Igs were positively correlated with each other. In brief, serum IgG at D1 was positively correlated with serum IgA at D1 (r = 0.58), D2 (r = 0.49), and D7 (r = 0.53) and in turn, serum IgA at D1 and D2 was positively correlated with salivary IgG at D2 (r = 0.52 and 0.55, respectively). In conclusion, there is an association between serum and salivary Igs in newborn calves. The absence of a statistical significant correlations between serum and salivary IgG and the low to medium correlations between serum and salivary Igs could be related to the heterogeneity of the transferred immunoglobulin mass per unit of BW that occurred in this study, to differences in the salivary flow rate between calves and to a possible time-dependent secretion of immunoglobulins from serum to saliva.

Session 4

Poster 18

Salivary immunoglobulin concentrations (IgG, IgM, and IgA) in calves during the first week of life

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Not much is known about the concentration of the immunoglobulins (Igs) in fluids other than blood in newborn calves. This study aimed to analyse the concentration of Igs in serum and saliva of calves during the first week of life. Saliva and blood samples were obtained from 20 calves at birth (D0) and at day 1, 2 and 7 after birth (D1, D2, and D7, respectively) to determine total protein (TP), IgG, IgA, and IgM concentrations in serum and saliva. A mean of 3.8±0.64 L of colostrum was administered within the first 6h after birth (96±73 min). Total protein in serum and saliva, and all Igs increased after colostrum intake, gradually decreasing in the following days. Salivary IgA was the highest at D0 (32.14 ug/mL) compared with other salivary Igs, showing the presence of mucosal immunity at a very early age in life. Salivary IgG concentration was the highest at D1 (104.95 ug/mL), decreasing rapidly at D2 (42.07 ug/mL). Salivary IgM concentration was lower compared to IgG or IgA. Nevertheless, it had a significant increase at D1 (5.21 ug/mL).

Impact of diarrhea and respiratory diseases in calves on reproductive performance in dairy breed heifers
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The most prevalent diagnoses in calves on dairy farms are diarrhea and respiratory diseases. These diseases not only have a negative impact on animal well-being but also have an economic impact for the dairy farms, resulting in treatment costs, retarded growth and increased time attending to sick calves. The occurrence of health problems early in life and during the rearing period might subsequently have an impact on performance, e.g. development and reproduction. This study aims to determine the influence of calf diseases at the age of first breeding. Therefore, data from Holstein Friesian and Normande calves from birth to the age of first artificial insemination (AI) were examined. More than 79,000 recorded health events were reported between 2005 and 2024 from 2,829 herds located in the Northwest of France. Health data were transmitted to INNOVAL and SYNETICS from the GENOSANTE network. A total of 10 investigated symptoms were grouped in diarrheal illness (diarrhea, neonatal diarrhea, cryptosporidiosis, coccidiosis), respiratory disease (flu, cough, bronchopneumonia, pneumonia) and umbilical hernia. In calves up to 2 months of age, diarrheal illnesses and umbilical hernia were the most prevalent diseases. Respiratory diseases occurred in all age groups but were particularly influenced by seasonal effects. The latest results on the impact of respiratory and diarrheal diseases in early calf life on age at first AI will be presented.

Pilot study to identify stress level at cow-calf separation after a few days of contact

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Calf separation from its mother at birth represents a hot topic due to the implications on public opinion. In 2023 the EFSA report indicated to keep the calf with the dam for a minimum of 1 day after partum. It is necessary to understand whether a short period of contact has positive or negative effects on calves and cow, in terms of separation stress. Cortisol hormone can be a good indicator of physiologic status after a potential stressful event. Three calves and two cows were selected to identify basal cortisol level during no stressful condition. At the third days after birth, during which cows and calves were kept together, saliva samples were collected from 11 dam and 18 female calves, 20 minutes after the separation. Hair samples were collected from different calves' bodies areas, to identify the variability of cortisol concentration depending on body region. An enzyme immunoassay (SALIMETRICS ASSAY #1-3002) was performed to assess cortisol concentration in saliva samples and hair, after an extraction procedure. The identified basal level of cortisol was 0.10 µg/dl for cows and 0.20 µg/dl for calves. The average cortisol concentration in saliva was 0.13 ±0.06µg/dl for cows and 0.19 ±0.22µg/dl for calves, median value was 0.12 µg/dl for cows and 0.11 µg/dl for calves. The high variability for cortisol level of calves shows individual physiological effect, particularly for young animals even though separation a few days after birth seems to be non-stressful. The cows' samples showed numerous values above the basal cortisol level, however, the values were not high and probably attributable to separation procedures. The hair of calves showed high variability in the cortisol concentration of the different body regions, with tail and nose having higher cortisol concentrations than the dorsal zone.

Keeping up with changing standards: A survey on how Canadian farmers currently rear dairy calves

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Decades of research has helped inform 'best practices' on how to care for dairy calves, but little is known about how these practices are adopted on commercial farms. Moreover, new regulations regarding the care of dairy cattle in Canada will require changes to calf rearing management on farms. The aims of this study were to describe the rearing practices of dairy calves and the methods used to assess calf growth on farms in British Columbia (BC), Canada. All 437 dairy farms in the province were invited to participate in an online survey distributed from June-December 2023. A total of 63 responses were received (representing 14.4% of the farms in BC). The average milking herd size was 166.7 + 172.1 cows, and the primary breed was Holstein (84.1%). Participants reported that on average 2.8 + 1.5 employees were responsible for the daily care of pre-weaned calves. Our results showed that 63.5% of participants housed calves individually, 25.4% socially housed calves in groups of two or more and, 11.1% of farms housed calves with a combination of individual and social housing pre weaning. The mean maximum milk allowance offered was 9.4 + 2.8 L/d with 87.1% offering >8 L/d. Teat feeding was used on 71.7% of farms with 16.3% using automated milk feeding systems and two responses reported feeding calves via the dam or nurse cows. Weaning age averaged 75.8+16.3 d, with calf age being the primary indicator to determine weaning. About half (52.4%) of farms reported measuring pre-weaning growth, where 31.7% of farms reported to have a target growth rate, only one participant stated that they had access to a scale. These results suggest that dairy farms in BC are following science-based recommendations by providing calves with increased milk rations. In addition, some farms have adopted social housing, albeit further research is needed to facilitate the transition to social housing by the 2031 regulatory deadline. Finally, our findings underscore the considerable room for improvement in monitoring calf growth; improved tracking of calf performance is likely to help farmers make better weaning decisions, including when and how to wean calves from milk onto solid feed.

Session 4

Poster 22

Enriching colostrum IgG concentration by ultrafiltration has no negative effects on in vitro cell viability and wound healing capacity

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An insufficient availability of high-quality colostrum contributes to a high prevalence of failure in passive transfer of IgG in calves, having negative impacts on calf health. By vibration-shear ultrafiltration technology, the IgG concentration in low-quality colostrum can reach high-quality levels (i.e., IgG > 50 g/L). However, ultrafiltration of colostrum carries the potential risk of compromising its bioactivity, consequently impacting the development of the juvenile intestine and its capacity of wound healing. The objective was to assess the bioactivity of non-processed colostrum (NPC) and colostrum enriched in IgG through ultrafiltration at 5 (UFC-5) or 30 kDa (UFC-30) molecular weight cut offs. This was accomplished by evaluating their impact on cell viability and wound healing capacity by using a fetal small intestinal cell line model. A total of six colostrum samples of each were included. Cells were treated with whey fractions of NPC, UFC-5, and UFC-30. Cell viability and wound healing capacity were evaluated by alamarBlue™ assay and xCELLigence real time technology, respectively. The effects were analysed by a linear mixed model with colostrum treatment, whey concentrations and their interactions as fixed effects and concentration nested within treatment nested within samples and assays as random effects. The UFC-5 showed higher cell viability than UFC-30 (201 vs 164%), with no difference observed between NPC and UFC. All colostrum treatments stimulated wound healing, with no difference between treatments. Considering this, it would be preferable to use a 5 kDa rather than a 30kDa MW cut-off for IgG-enrichment of colostrum by ultrafiltration.

Saccharomyces cerevisiae fermentation products supplementation to dairy calves: effects on growth, metabolism, and immune status

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To evaluate the effect of supplementing *Saccharomyces cerevisiae* fermentation products (SCFP) on calf growth, metabolism, and immune status, 18 Holstein female calves were allocated to 2 treatments: control (CTR; n=9; no supplementation) or supplemented group (SCFP; n=9; 1 g/d of SmartCare®, Diamond V, in milk replacer plus 5 g/d of NutriTek®, Diamond V, from 5 to 70 d in the starter). Calves were weaned at d 60 and feed intake was recorded up to d 70. Body weight (BW) was measured until d 160, and blood samples were collected regularly until d 70 for the assessments of metabolic profile, plasma volatile fatty acids, and polymorphonuclear cells (PMN) phagocytosis. Data were analyzed using a mixed model for repeated measures. Compared with CTR, SCFP had increased post-weaning growth, with greater average daily gain from d 70 to 100 (+57%; P<0.01) and BW from d 100 to 160 (~9 kg more; P<0.04). SCFP tended to have greater plasma β -hydroxybutyrate at d 60 (P=0.06) and urea at d 70 (P=0.08) and had higher plasma concentration of acetate and propionate (P<0.05), suggesting an increased rumen activity. The latter might reduce weaning stress. Plasma myeloperoxidase was higher in SCFP at d 70 (P=0.04), as well as PMN phagocytosis at d 60 and 70 (P=0.03), indicating improved immune system functionality. In conclusion, supplementing SCFP improved the metabolic and immune status of dairy calves and supported the post-weaning growth.

Session 4

Poster 24

Effect of age, undernutrition and hydroxytyrosol supplementation on metabolic stress markers during the last third of gestation in beef cattle

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Undernutrition in late pregnancy is a common scenario in beef herds. Hydroxytyrosol (HT), a polyphenol present in olive leaves, has been shown to improve antioxidant capacity. The aim of this study was to assess the effects of undernutrition (100% vs. 60% of requirements) and HT supplementation (Control vs. HT, 178±36 mg/kg) during the last term of pregnancy on whole blood gene expression of redox and energy metabolism markers in beef cattle. Blood samples were collected from 56 cows at week 37 after mating. Gene expression of SOD1, CAT, GPX1, NRF2, ALOX5, SCD, and IGF-R was analysed by qPCR. A general linear model including fixed effects (nutrition, HT supplementation and cow age (<8 vs. ≥8 years)) and their interactions was used to compare gene expression with JMPPro software. A higher SCD expression was observed in 100% than 60%-cows, while higher ALOX5 expression was observed in HT than control cows (P < 0.05). Lower SCD expression would indicate impaired lipogenic activity in 60%-cows, whereas ALOX 5 upregulation could imply the activation of a prompt termination of the inflammatory stress during the peripartum in HT cows. In <8-year-old cows, there was an increase in NRF2 and IGF-R expression, along with a decrease SOD1, CAT, and GPX1 expression compared to ≥8-year-old cows (P < 0.05). Our results suggest that older cows exhibit higher metabolic flexibility and higher antioxidant status than younger cows during peripartum period. Overall, cow age, undernutrition and HT supplementation influenced metabolic stress markers during peripartum period in beef cattle. Funded by PID2020-113617RR-C21/22.

Interesting genes and effect of inbreeding depression over traits with economic and productive importance in Rubia Galega beef cattle breed

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The Rubia Gallega (RG) is a Spanish autochthonous beef cattle breed primarily found in the autonomous region of Galicia. Previous research has identified a remarkable inbreeding depression effect linked to economically important traits. This study seeks to build upon prior research by examining the association between inbreeding effects on four economically significant traits (Calving Interval – CI -, Age at First Calving – AFC -, Birth Weight – BW -, and Weight at 210 days –W210-) with the presence or absence of Runs of Homozygosity (ROHs) across the autosomal chromosomes. The goal is to deepen our understanding of the consequences of inbreeding in the RG population and its impact on these traits, as well as to identify genes or genomic regions linked to desirable or undesirable inbreeding effects. The data set consist of genotypes with 57,490 single nucleotide polymorphisms (SNPs) from 4,878 individuals. All of them are recorded for BW, and we dispose of phenotypic data from 3,503, 3,315 and 3,285 of them for AFC, CI and W210, respectively. ROHs were identified utilizing the “consecutiveRUNS.run” function of the R package “detectRUNS,” with parameters categorized by length (in Mb): [1, 2], (2, 4], (4, 8], (8, 16], and >16. Subsequently, a matrix was created indicating whether each SNP was situated within an ROH for each animal. A t-test was then conducted to assess the significance of the effect of SNP presence within an ROH, following BLUP analysis performed by the BLUPF90+ software. A total of thirteen regions within BTA were identified as significantly associated with inbreeding effects. Notably, these include BW (2, 3, 6, 11, 13, 28), AFC (1, 2, 4, 5, 7), and W210 (2, 14, 21 – two regions -), while no significant regions were observed in IC. Among the genes located within them, the MSTN (Myostatin), CREB5 (CAMP Responsive Element Binding Protein 5) and FABP4 (Fatty Acid Binding Protein 4) can be highlighted.

Session 4

Poster 26

Bovine faecal biomarkers of intestinal inflammatory process: calprotectin and lactoferrin, a comparative study

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Inflammation of the gastrointestinal tract impairs its functionality causing diarrheas that compromise calves' health and growth. In inflammatory conditions, calprotectin and lactoferrin are released to the intestinal lumen and their determination in faeces is used as biomarkers of inflammation in the gastrointestinal tract. The objectives of the present work were: a) validation of an ELISA assay developed for the measurement of lactoferrin in milk for its use in calves' faeces, b) study the influence of the water content of faeces on fecal calprotectin (fCAL), previously validated for calves, and fecal lactoferrin (fLF), and c) correlation between fLF and fCAL. The analytical validation was performed following the ASVCP Guidelines. Spearman's correlation coefficient was used to evaluate the influence of faecal water content and correlation between both biomarkers. Analytical validation achieved good results for both faecal biomarkers. A very strong correlation was observed between fCAL concentration in fresh and dry faeces ($r = 0.903$, $p < 0.01$) while a moderate correlation was observed for fLF ($r = 0.648$, $p < 0.01$). Correlation between both biomarkers was moderate in fresh faeces ($r = 0.514$, $p < 0.01$) and in dry faeces ($r = 0.561$, $p < 0.01$). In conclusion, a) the lactoferrin ELISA kit is valid for its use with calf faecal samples, b) fLF concentration is more influenced by faeces moisture than fCAL concentration, and c) both proteins can be proposed as good biomarkers of intestinal inflammation, since analytical procedures were satisfactory.

Effects of goat willow's tannin profile on in vitro methane production and rumen fermentation

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This study assessed the effect of high-procyanidin (HPC) and high-prodelphinidin (HPD) goat willow (GW) on in vitro gas, methane (CH₄) and volatile fatty acid (VFA) production when included at 20% DM basis. Diets included grass silage (GS), HPC (96% PC in tannins), HPD (73% PD in tannins); with the addition of polyethylene glycol (+PEG; 1.7:1 tannin:PEG) or without. Diets were incubated for 72h at rumen fluid:medium at 1:9 v/v in triplicate. Gas pressure (psi transducer) and CH₄ concentration of gas (gas chromatography) were measured throughout. DM degradability (DMD) and VFA concentrations were assessed at 72h. Data were analysed by linear mixed models using treatment as fixed factor and batch run (1-3) as random factor. HPC, HPD, HPC+PEG, HDP+PEG diets had lower (P<0.001) DMD (697, 683, 677, 680, g/kg DM), and 72h production of cumulative gas (120, 113, 144, 133 ml) and CH₄ (9.1, 9.1, 10.6, 11.0 ml), than GS (754 g/kg DM, 163.7, 15.7 ml). GS produced more (P<0.001) VFA and acetic acid (70.0, 42.2 mM) than the four GW diets (60.4-67.8, 35.2-39.7 mM respectively), and less (P<0.001) butyric acid (4.5 mM) than HPD (7.7 mM). Adding GW reduced in vitro CH₄ production, partly due to tannins.

Session 5

Theatre 2

Effects of ten freshwater microalgae on in vitro ruminal methane production and digestibility

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This study aimed to evaluate the effects of 10 freshwater microalgae, namely, *Auxenochlorella protothecoides* (Ap), *Chlamydomonas pulvinata* (Cp), *Chlorella luteoviridis* (Cl), *Chlorella variabilis* (Cv), *Euglena mutabilis* (Em), *Parachlorella kessleri* (Pk), *Scenedesmus acuminatus* (Sa), *Stichococcus bacillaris* (Sb), *Tetradesmus obliquus* (To) and *Tetraselmis gracilis* (Tg), on in vitro ruminal methane (CH₄) production and nutrient digestibility using Hohenheim Gas Test. Microalgae were cultured in CO₂ incubator at 2% CO₂, light at 115 mmol/m².s in a 16:8 day/night cycle at optimal temperature of each strains. The Ap, Cp, Cl, Cv, Em, Pk, Sa, Sb, To and Tg possess crude protein contents of 17.2, 46.3, 40.2, 44.0, 42.5, 42.5, 42.3, 38.5, 37.0, 61.0 and 27.0%DM, a PUFA content of 59.0, 37.3, 69.1, 38.0, 42.3, 55.5, 40.5, 48.7, 35.3 and 33.2% of total fatty acids, respectively. The microalgae were tested at 15% replacement in a basal diet (Control) of 40%DM grass silage, 40% maize silage, 15% hay and 5% concentrate. Data was analyzed in mixed model in R. Ruminal CH₄ production was reduced by 15.4, 13.5, 17.4, 11.3, 9.70, 16.4, 10.0, 13.5, 11.2 and 12.2% (P < 0.05) for Ap, Cp, Cl, Cv, Em, Pk, Sa, Sb, To and Tg, respectively, compared to Control. In vitro organic matter digestibility was reduced by 3.31, 5.17, 5.14, 3.44, 2.67, 5.37, 2.76, 4.87, 2.55 and 3.29%pt (P < 0.05) for Ap, Cp, Cl, Cv, Em, Pk, Sa, Sb, To and Tg, respectively. Incorporating microalgae in ruminant diets has the potential to reduce enteric CH₄ emissions, further investigation is needed.

In vitro rumen fermentation characteristics of insect processed proteins compared to a conventional protein source

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This research aimed to evaluate the nutritional composition, in vitro digestibility, and gas production kinetics of Black Soldier Fly and *Tenebrio molitor* processed proteins compared with soybean meal f.e. as a reference raw material. Nutritional characterization and in vitro ruminal fermentation tests were performed to determine in vitro short-chain fatty acids production, the gas production profile and methane production. Black Soldier Fly (BSF) and *Tenebrio molitor* (TM) were both full fat meal with a mean protein content of 41 and 61 % respectively and the soybean meal had 45 % of crude protein. Total gas production (value expressed in mL as mean \pm SD) was lower for both Black Soldier Fly (27 \pm 4.5) and *Tenebrio molitor* (23 \pm 3.3) processed proteins compared to soybean meal (77 \pm 2.1). Similarly, differences have been observed in the case of short-chain fatty acids production which was higher when soybean meal was used as substrate than with BSF and TM (4398, 1827 and 1676 μ mol, respectively). However, when considering fatty acid molar proportion differences were observed only in case of propionate, isobutirate, isovalerate, and valerate. Interestingly methane production (value expressed in μ mol as mean \pm SD) was lower for insect processed proteins (BSF 70 \pm 15; TM 59 \pm 4.5) compared to soybean meal (282 \pm 7.6). Obtained results indicated that insect processed proteins hereby tested were characterized by low fermentation profile, thus it might speculate that they could represent a valid by pass protein source.

Screening of Mediterranean agro-industrial by-products on in vitro ruminal fermentation

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The utilization of polyphenol-rich agro-industrial by-products for ruminant diet would be a sustainable and circular option to mitigate pollutant emissions. In this study, 16 by-products sourced from Greece, Italy, and France were evaluated at 10% inclusion level in a basal diet using an in vitro ruminal fermentation assay. Dry matter degradability (DMD), gas production (CH₄, CO₂), volatile fatty acids (VFAs), and ammonia (NH₃) concentrations were monitored. A mixed-model ANOVA test was utilized to assess the effects of each by-product relative to one another. Additionally, a Dunnett post hoc test was conducted to compare the by-products against the control, comprised solely of the basal diet. Results revealed significant compared to control diet reductions in CH₄ production with French grape, Italian hazelnut, and Greek pomegranate by-products ($p < 0.001$). French grape, Italian and Greek pomegranate led to an increased CO₂/CH₄ ratio ($p < 0.01$) and decreased NH₃ production ($p < 0.05$) compared to control diet. In addition, the French grape by-product significantly reduced DMD ($p < 0.05$), without affecting VFAs production. Furthermore, this by-product demonstrates a decrease in CH₄ and NH₃ production ($p < 0.001$) compared to the control. These findings suggest a shift in fermentative pathways from CH₄ to CO₂, affecting methanogenic microorganisms. Despite the lower DMD observed, the French grape by-product emerges as the best candidate for mitigating pollutant emissions.

Effect of the use of *Lactiplantibacillus plantarum* strain 5BG and of a commercial mix of polygalacturonase and glucanase on the quality of artichoke bracts silage

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The study aims to evaluate the viability of ensiling artichoke bracts by the addition of a bacterial strain, *Lactiplantibacillus plantarum* 5BG(LP), and a commercial mix of polygalacturonase and glucanase, Vinotaste®(PGE). The artichoke bracts (AB) were obtained in a unique batch and split in 10 subsamples. Five experimental treatments were applied (2 subsamples per treatment) i) 10⁸ cfu of LP per g of AB(5BG); ii) 10⁸ cfu of LP per g and 1.6 g of PGE per kg of AB (5BG+E); iii) 1.6 g of PGE per kg of AB (E); iv) 10⁷ cfu of LP per g of AB(5BGL); v) 910 ml of distilled water (C). All the inocula were homogeneously sprinkled on each sample before ensiling. Each aliquot was compressed in plastic silos, hermetically closed, and stored at room temperature for thirty days. The fresh and ensiled AB were analyzed for chemical gross composition. In vitro digestion test (Daisy II, Ankom) for the apparent digestibility evaluation and in vitro gas production (Ankom Gas Analyzer) were carried out. Raw and ensiled samples were used also for microbiota analysis. The data set was processed by an ANOVA model, while metagenomics results were used for a PCA analysis. The 5BG, 5BG+E, and 5BGL theses led to a reduction in total gas production (P<0.05). Moreover, microbiota modifications during AB fermentation showed a clear cluster of the microbial diversity among the theses with LP than the ones without. Apparent digestibility was not affected by the treatments. The use of the LP strain showed to be an effective strategy for the production of AB silage, able in reducing enteric methane production without impairing apparent in vitro digestibility

Effect of conservation length on fermentation characteristics of teff as alternative forage for Mediterranean livestock farming systems

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This study aimed to investigate the silage process of teff (*Eragrostis tef*) as a C4 drought-resistant forage for Mediterranean livestock farming systems. Moxie® (Barenbrug Co.) seeds were sown in central Italy after the durum wheat harvest, and the forage was cut after 70 days at approximately 30% dry matter content. Following chopping to about 4 cm, the fresh forage was ensiled in 20 L plastic silos fitted with a gas-release-only lid. The silos were opened at 30 and 60 days. The silages were analysed for water activity, chemical profile, microbial count, and aerobic stability. The earlier opening allowed for the maintenance of higher water activity, dry matter, crude protein, fat, starch, and yeast content. A longer conservation period increased buffering capacity, ADL, non-structural carbohydrates, and *Lactobacillus* spp. The length of ensiling did not affect other parameters such as weight loss, pH, ash, crude fiber, NDF, ADF, aerobic stability, and mold content. The results of this experiment suggest that a shorter ensilage period (30 days) does not lead to qualitative deterioration, and key storage parameters (pH, aerobic stability) are not altered. Meanwhile, higher levels of crude protein, fat, and starch are maintained. Teff can be useful in the event of forage shortages, as it can be sown even after the main cultivation in mid-summer, can be ensiled when haying is at risk (late summer-early autumn), and it can be fed to animals after a short conservation period. Funding: This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022).

Understanding gastrointestinal tract transcriptomic adaptations in dairy cattle during lactation reveals molecular tissue and stage specific adaptations

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To accurately predict maintenance requirements in lactating dairy cattle it is necessary to consider the dynamics of visceral organ tissue mass and metabolism in response to stage of lactation as they have an outsized role in nutrient use efficiency. We used next-generation sequencing and transcriptomic profiling of rumen, duodenum, and colonic epithelia sampled throughout the lactational cycle (Dry and 3, 14, 28, 45, 120, 220, and 305 days in Milk). Biopsies collected from cows were subjected to global transcriptomic profiling to identify tissue and stage-specific gene expression, time series clustering of RNA-seq data, co-expression of gene network analysis, as well as tissue-specific expression patterns, functional enrichment and cell type deconvolution. Striking changes in immune function gene expression begin to occur in the rumen and colon tissues as soon as day 3. Duodenal gene expression changes are associated with both immune function as well as nutrient transport, which is consistent with the rapid increase in nutrient demands for the cow. In the transition period (d 14), all three tissues exhibited specific changes in nutrient transport-related gene expression. Using a serial sampling approach enabled direct comparison of gene expression patterns within and among tissues during different phases of lactation and lends insight into the tissue-specific characteristics of these important gastrointestinal tissues across the lactation cycle.

Session 5

Theatre 8

Nutritional value of *Nannochloropsis oceanica* for post-weaning piglets

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The identification of novel or underexploited feed resources to be used as alternatives to conventional feedstuffs constitutes an urgent matter towards increased sustainability of the animal feeding sector. Microalgae, like *Nannochloropsis oceanica* (NCO), due to its interesting composition, rich in protein, lipids, and minerals, could be a viable option for this, specifically for monogastric feeding. This study aimed to estimate the nutritional value of spray-dried NCO biomass to better understand the applicability of this microalga in weaned piglet feeding. Piglets were individually housed in metabolic cages and randomly assigned to 4 dietary treatments (n=6): control, 5, 10 and 15% dietary incorporation of NCO as a replacement of the basal diet. The experiment lasted two weeks (with a previous adaptation period of 4 days). Feed intake was equalized between diets and monitored daily. Faeces and urine were collected and weighed. Regression estimates of NCO total tract apparent digestibility (TTAD) values were obtained using a mixed model. The TTAD of dry matter (DM), crude protein (CP) and ether extract obtained were 725 ± 36.4, 711 ± 75.9 and 513 ± 83.9 g/kg, respectively. Digestible energy, metabolizable energy and digestible CP were calculated and its values were 12.7 MJ/kg DM, 12.4 MJ/kg DM and 177 g/kg DM, respectively. Further research should be targeted to understand how to use the NCO biomass properties in piglet nutrition.

Growth performance and carcass traits of heavy pigs fed diets with a partial to total replacement of soybean meal with spirulina powder

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We evaluated the effects of a partial to full replacement of soybean meal (SBM) with Spirulina in the diets for heavy pigs, as a novel protein source. The study involved 88 gilts and barrows, allotted into 4 groups (2 pens/group) which received a control diet (C) based on cereals and SBM or one of 3 diets formulated by replacing nearly 33% (SP1), 66% (SP2) or 100% (SP3) SBM with Spirulina. The feeds were isoenergetic, isoproteic and isoaminoacidic. A mild feed restriction was applied, and feed allowance increased from 2.3 kg at the start (52 ± 4 kg body weight – BW) to 3.2 kg/d at the end of the trial (175 ± 6 kg BW). Growth performance, feed intake, gain to feed ratio, and the weight of carcasses, loins, hams and lards have been analyzed according to a linear mixed model including the fixed effects of sex and diets and their interaction, and the pen within diet as random effect. Palatability of diets was not depressed by the inclusion of SP, and pigs of the different groups had very similar feed intake. Growth rate and gain to feed ratio averaged 894 ± 40 g/d and 0.341 ± 0.016 g/g, respectively, and growth parameters were comparable ($P > 0.10$) across groups. Also carcass and commercial cuts weights were unaffected by diet. Therefore, the replacement of SBM with spirulina did not exert any effect on growth and carcass traits of growing pigs. The effects of such a replacement on meat quality should be addressed. This study was carried out within the Agritech National Research Center and funded by European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022).

Soybean meal fermentation: is it worth investigating in Europe? A review

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From 2020 to 2023, we systematically reviewed soybean meal processing to enhance animal feed quality for land livestock (ruminants, pigs, poultry). Fermentation emerged as a promising method. Of the total of 49 articles identified, 42 were from Asia (China (30), South-Korea (3), Taiwan (3), Iran (3), Thailand (1), Sri Lanka (1), Saudi Arabia (1)), 3 from Europe (Poland (2), Denmark (1)) and America (USA (2), Mexico (1)), and 1 from Africa (Egypt). Fermented soybean meal (FSBM) was mainly (32 articles) evaluated in diets for weaned piglets and broilers. FSBM was mainly produced by solid-state fermentation (with 1 to 3 fermentation steps). Bacillus and Lactobacillus bacteria, often combined, were mainly used. There was a wide range of experimental protocols: each article had its own process for producing FSBM, and the inclusion rates of FSBM in the diets varied from 2 to 35%. The authors tested the inclusion of FSBM as a full/partial replacement of conventional soybean meal (CSBM) or animal protein diet to assess its alleged positive effects on diet composition, ANFs content, nutritional value, animal performance, health, microbiota, and quality of animal products. However, few papers tested the environmental and economic impact of FSBM diets. The conclusions of the articles were by far and large positive, confirming the benefits of FSBM over CSBM and animal-based diets for all livestock species. The positive effects were more noticeable in pigs than in poultry: in pigs, FSBM improved health, performance, nutritive value and product quality in 87%, 83%, 82% and 82% of the tests respectively, while these numbers were lower in poultry (75%, 82%, 57% and 61%). These results are certainly promising and it would be valuable to explore the potential of FSBM in European contexts, focusing on its nutritional and performance benefits, and also on its economic and environmental impact. The value of FSBM in ruminants, which has been little studied in Asian articles, could be further explored, particularly by considering strains of micro-organisms that are more cellulolytic for fermentation.

Extruded or Enzyme-Treated *Chlorella vulgaris* in Broilers Diets: Effects on Performance and Digesta Viscosity
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This study explores the impact of incorporating microalga *Chlorella vulgaris* (MCV), an alternative protein source to soybean meal, into broiler diets either untreated, enzyme-supplemented, or extruded, on animal performance and digesta viscosity. One hundred and twenty male Ross 308 broilers, from 7 to 35 days of age, were divided into four dietary treatments with 10 replicates each: CTR (maize and SBM), CV (20% MCV), CVenz (CV diet + 0.3% pancreatin) and CVExt (20% extruded MCV), with ad libitum feeding for 28 days. Performance parameters were determined and, at day 35, organ sizes, digesta viscosity and carcass yield were measured. All data was analysed with ANOVA, using SAS software. Key findings include that feed intake and average daily gain reduced across all MCV diets compared to CTR, yet CVenz and CVExt diets improved carcass yield relative to the CTR. CVenz diet yielded body weights and feed conversion ratios comparable to CTR, underscoring its potential for maintaining growth performance despite high levels of MCV inclusion. Digesta viscosity was not significantly affected by diet, suggesting the physical and enzymatic treatment methods employed did not detrimentally impact gut health. Organ weights and lengths varied with treatment, with CVenz and CVExt diets resulting in lower liver weights and changes in gastrointestinal tract lengths, indicative of physiological adaptations to the diets. Overall, this research highlights the feasibility of using enzyme-treated MCV as a sustainable protein source in broiler diets without compromising performance or gut health.

Session 5

Poster 12

Evaluation of different algae for ruminant feeding

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Algae are microorganisms that can be used as a source of nutrients in ruminant feeding. However, evaluation of microalgae can be challenging due to its microscopic size. The aim of this work was to evaluate the in vitro dry matter (DM) degradability (IVDMD) of various algae using ANKOM F58 bags. The chemical composition of five microalgae (*Tetraselmis* spp. (TE), *Tisochrysis lutea* (TI), *Scenedesmus almeriensis* (SC), *Nannochloropsis gaditana* (NA) and *Arthrospira* spp. (AR)) and one macroalgae (*Ulva* spp. (UL)) was analyzed. Three F58 bags with each algae sample were incubated with buffered sheep ruminal fluid from 3 sheep for 12 h to determine the IVDMD. In addition, algae were incubated in flasks for 12 h to measure gas production. The nitrogen (N) content was 70.9, 66.3, 96.2, 67.1, 110 and 68.6 g/kg DM, the ether extract (EE) content was 172, 268, 151, 122, 159 and 143 g/kg DM, and the ash content was 288, 185, 66.5, 206, 78.1 and 170 g/kg DM for TE, TI, SC, NA, AR and UL, respectively. The gas produced after 12h was 31.3, 18.8, 22.4, 42.7, 65.5, and 22.8 mL/g DM, and the IVDMD was 26.5, 47.4, 34.8, 41.0, 58.7, 37.3% for TE, TI, SC, NA, AR and UL, respectively. The IVDMD significantly and positively correlated with gas production ($r=0.61$; $n=16$). Some feed fractions are degraded and produce amounts of gas during in vitro trials. Adjusting this gas production for N, EE and ash content through linear regression, the correlation with the IVDMD was close to 1 ($r=0.98$). These results suggest F58 bags can be used to evaluate algae.

In vitro evaluation of discarded carrot and pepper as feed ingredients for ruminants

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Spain leads as the top producer of fruits and vegetables in the EU. Discards and leftover materials from harvesting and processing processes represent a significant part of this production and, after nutritive evaluation, they could be incorporated into the diet of ruminants. Chemical composition and in vitro rumen fermentation characteristics of carrot (CAR) and pepper (PEP) were assessed in this study. 0.5 g samples of both vegetables and barley as reference feed were placed into serum bottles containing 50 mL of buffered ruminal fluid and incubated for 24 h at 39°C. Total gas production, volatile fatty acids (VFA) and CH₄ production and pH were measured. Chemical composition of CAR, PEP and barley was, respectively, 10.1, 9.56 and 94.6 % DM (% of fresh matter); 92.9, 92.1 and 98.4 % OM, 8.08, 12.7, and 12.2 % CP, 0.79, 2.94 and 1.60 % EE, 41.6, 34.1 and 3.05 % sugars, and 12.4, 15.6 and 12.5 % NDF (% DM). Total gas production was significantly higher for CAR (P<0.05) compared to PEP and barley (96.5, 88.2 and 90.1 mL/bottle, respectively). Total VFA production was significantly higher for CAR than barley (4688 and 3920 µmol/bottle; P<0.05) with intermediate values for PEP (4378 µmol/bottle). Acetate molar proportion was similar for all substrates (P>0.05) but molar proportion of propionate was higher for CAR and PEP compared to barley (P<0.05). Consequently, acetate/propionate ratio was lower in CAR and PEP than in barley (1.97, 2.13 and 2.77, respectively). Methane production and methane/VFA ratio were not significantly different between substrates (P>0.05), with a methane production of 339.5, 335.7 and 304.0 µmol/bottle and a methane/VFA ratio of 0.073, 0.077 and 0.078 mol/mol for CAR, PEP and barley, respectively. Both vegetables appear to be an alternative in ruminant feed, replacing traditional feed such as barley in the ration. Further studies are needed to assess the effective inclusion of these vegetables into ruminant diets. Funding from the Spanish State Research Agency (AEI) and the European Regional Development Fund (Project PID2021-124648OB-C22) is gratefully acknowledged

Session 5

Poster 14

In vitro evaluation of discarded leek as feed ingredients for ruminants

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Sulfur compounds present in plants may modulate ruminal fermentation and some of them appear to inhibit methane production. Chemical composition and in vitro rumen fermentation characteristics of discarded leek (LEE), a vegetable rich in sulfur compounds, were assessed in this study, compared to a reference feed (alfalfa hay, ALF). Samples (0.5 g dry matter) of substrates were incubated in serum bottle with 50 mL of buffered ruminal fluid for 24 h at 39°C. Total gas production, volatile fatty acids (VFA) and methane production and pH were measured. Chemical composition of LEE and ALF was 7.65 and 95.8 % dry matter (% of fresh matter); 87.68 and 92.27 % organic matter; 18.29 and 20.48 % crude protein; 1.77 and 1.37 % fat; 19.20 and 7.1 % sugars; 22.56 and 31.56 % neutral detergent fiber; 18.31 and 22.98 % acid detergent fiber; 4.53 and 4.94 % lignin (% of dry matter). Total gas production was significantly different, being higher with LEE than ALF (74.3 and 59.5 mL/bottle). Total VFA production was significantly higher for LEE compared to ALF (3825 and 3275 µmol/bottle). No differences were found in molar proportions of acetate, propionate and butyrate (P>0.05) and consequently acetate/propionate ratio was similar for both substrates (2.72 and 3.12 for LEE and ALF, respectively). There was no difference in methane production between LEE and ALF (178 and 145 µmol/bottle respectively). Additionally, CH₄/VFA ratio was similar for both substrates (0.046 and 0.044 mol/mol, respectively). In conclusion, LEE seems to be an alternative to be included as feed ingredient in ruminant diets although no effect in CH₄ production was observed when compared in vitro with alfalfa hay. Additional research is required to assess the optimal incorporation of this vegetable into ruminant diets. Funding from the Spanish State Research Agency (AEI) and the European Regional Development Fund (Project PID2021-124648OB-C22) is gratefully acknowledged

Use of sunn hemp as forage for beef cattle

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Sunn hemp (*Crotalaria juncea* L.) is a warm-season annual legume and has been widely used as cover crop in the world. Livestock producers have interest in using sunn hemp as forage due to superior biomass accumulation and nutritive value; however, sunn hemp has pyrrolizidine alkaloids, which may negatively affect forage quality. The experiment was conducted at the UF/IFAS Range Cattle Research and Education Center, Ona, FL from April to May 2021. Treatments were calves receiving sunn hemp or stargrass (*Cynodon nlemfuensis* Vanderyst) hay distributed in a randomized complete block design with 12 replicates. Calves were supplemented with concentrate at 1.3% BW daily (18% CP and 78% TDN). Twenty-four Angus crossbred calves were weaned with 90±14 kg BW and grazed annual ryegrass (*Lolium multiflorum* Lam) with 1% BW concentrate supplementation for approximately 60 d. Subsequently, calves were randomly distributed and assigned to experimental units (pen) with initial BW of 184±10 kg. ‘Crescent Sunn’ sunn hemp hay was harvested approximately 49 d after seeding when 75% of the plants were initially flowering. The average concentration of pyrrolizidine alkaloids from sunn hemp was 58 µg/g DM. Early-weaned calves receiving stargrass had greater ADG than sunn hemp (0.5 vs. 0.4 lb/d), and had greater forage DM intake (1.2 vs. 1.0% BW) and total DM intake (2.5 vs. 2.3% BW). Conversely, diets with sunn hemp had greater *in vivo* DM digestibility than stargrass (68 vs. 62%). There was no difference between treatments on parasite egg count (mean = 45 eggs/3 g feces) and coccidia egg count (1 log10 eggs/g feces). Early-weaned calves receiving sunn hemp consumed 0.25 g pyrrolizidine alkaloids/d; however, there was no detectable amount (<0.05 µg/g DM) of pyrrolizidine alkaloids in the feces. The presence of pyrrolizidine alkaloids in sunn hemp may be the potential cause of decreased early-weaned calves forage intake and animal performance.

Session 5

Poster 16

Assessment of the potential of BSF meal in broiler chicken feed

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The use of insect meal in animal feed is an opportunity to reduce the pressure on the production systems of raw materials for animal feed, especially of protein sources, significantly reducing the “Food vs Feed” competition and leveraging a new sector with great potential. This study is being conducted under the Mobilizing Agenda InsectEra, project number AAC n°02/C05-i01/2022. The aim of this study is to evaluate the added value of BSF meal in broiler feed, considering zootechnical performance and animal health. Commercial broilers (n=320) were allocated to four experimental groups in 2 stages of animal growth (phase 1: P1 and phase 2: P2) according to the following: 1 control C) and three treatments (T1, T2 & T3) considering 2 different insect meal incorporation rates according to the following: T1 – 2,5% (P1), 2,5% (P2); T2 – 5% (P1), 5% (P2) and T3 – 5% (P1), 2,5% (P2). The broilers were housed in experimental pavilions that mimic industrial ones, respecting all current welfare standards, namely area per animal, temperature, light intensity and photoperiod. Each pavilion had automatic drinking fountains and a feeder for permanent access to water and food, respectively. Each experimental treatment had 4 replicas (pavilions) and each pavilion had 20 animals. Measurements were made at the beginning, at day 18, when the feed was changed (phase 1 to phase 2), and at the end of the trial. The parameters measured were: Individual weight of the birds; average daily weight gain; average daily feed intake per bird on a weekly basis; average daily intake; feed conversion ratio; mortality and morbidity. The intestinal health of the animals was also assessed by measuring the length of caecae, small intestine and its sections and the size of the intestinal villi.

Intercropping cash and forage crops to improve the land equivalent ratio of Mediterranean organic cereal-based systems

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This work was part of the joint research project “RE-FARM”, whose main objective is to combine regenerative agriculture methods with proximal sensing to enhance productivity, crop quality, and ecosystem services of wheat-based farming systems in south Italy and Texas, USA. In the first year (2023), a field trial was set-up in an organic farm located in Sicily, Italy. Durum wheat (*Triticum durum* Desf.) sole crop (control) was compared with durum wheat intercropped with forage legumes, with the aim to reduce weed pressure and nitrogen requirement, to increase grain yield and protein content, to extend the biological cycle of legume species for forage production and to improve the land equivalent ratio (LER). Three Mediterranean forage legumes (*Trifolium subterraneum* L., *Medicago polymorpha* L., and *Lotus corniculatus* L.), were selected based on predominant winter growth, different growth habits and life cycle length. Focusing on forage performances, results indicated that intercropped forage species differed in term of dry matter (DM) yield, DM content at harvest and biological cycle length. At the wheat harvest in July, the DM yield was significantly higher in *T. subterraneum* than *M. polymorpha* and *L. corniculatus* (1.65 Mg ha⁻¹ vs 0.35 and 0.30 Mg ha⁻¹, respectively). Accordingly, intercropping allowed to improve the LERs, which were higher than one (i.e., that of the *T. durum* sole crop). The DM content was the highest in *M. polymorpha*, as it reached senescence phase already in the second decade of June. *T. subterraneum* had a DM content of 51±4.9% as it did not complete the cycle at harvest. *L. corniculatus* was still at the flowering at the moment of wheat harvest, and its DM was only 21.8±1.9%. Overall, this study showed that intercropping can be a suitable agroecological practice to enhance the LER and to provide both forage and grains from organic cereal-based systems. Acknowledgements: This work was supported in part by the Italian Ministry of Foreign Affairs and International Cooperation”, grant number US23GR18 (RE-FARM project, CUP: J43C23000110001).

Nutritional quality of feed resources used by smallholder dairy farmers in the Northern Province of Rwanda
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Improving the nutritional quality of dairy cow diets in Sub-Saharan Africa can uplift small-scale farmers' livelihoods. This study aimed to assess the nutritional quality of feeds used by smallholder farmers (SHF) in the Northern Province of Rwanda through chemical analyses and in vitro gas production. A total of 218 feed samples were collected from 178 households. Twenty different feeds were identified and classified as cultivated forages (CF, 4), natural pastures (NP, 5), crop residues (CR, 9), and fodder trees (FT, 2). The most utilized feed resources were NP (51%) and CF (46%), followed by CR (23%) and FT (2%). *Pennisetum purpureum*, and assorted grass were the prevalent feeds used by the SHF (42%, and 32% respectively). Crude protein (CP) and fibre (NDF) contents were on average 10.5±1.2 and 53.6±12.5 in CF, 9.4±1.8 and 57.0±8.6 in NP, 11.1±4.2 and 53.7±12.5 in CR, and 29.7±7.2 and 40.1±13.7 in FT, respectively. Among all feeds, FT as *Sambucus nigra* had the highest content of CP (34.8%), and metabolisable energy (ME; 10.7 MJ/kg DM), compare to crop residue in banana pseudo-stem (CP; 4.4% and ME; 5.0 MJ/kg DM). *S. nigra* and *Calliandra calothyrsus* (24.6% CP) could be good supplementary feeds for those with lower nutritional quality. This study contributes to defining possible interventions and strategies to improve feed management practices, enhancing productivity and resilience of SHF in Rwanda. The study is part of the “Undernutrition” project, funded by the Swedish International Development Cooperation Agency in partnership with the University of Rwanda.

Cu, Zn, and Mn sources effect on 48 hour in-vitro fermentation and CH₄ production

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The objective of the trial was to evaluate copper (Cu), zinc (Zn), and manganese (Mn) mixtures from various sources on 48 hour in-vitro fermentation. Supplemental trace minerals (STM) calculated to be 125 Cu, 750 Zn, and 500 Mn (mg/d), for a lactating dairy cow consuming 25 kg DM with a 120 L rumen volume and scaled to fit a 250 mL vessel. Treatments were split into two runs with 6 replicates per treatment: Run 1: Control (C1, no trace mineral), Sulfate (SO₄), Selko® IntelliBond® (IB), Nutrilock® Chemlock (NC), Phibro Vistore™ (PV). Run 2: Control (C2, no trace mineral), SAM Nutrition (SN), Orffa Excential Smart (OE), and Ecotrace Glycinate (EG). Data were analyzed as a complete randomized design with PROC MIXED in SAS (SAS Institute Inc, Cary, NC). Significance equaled $P \leq 0.05$ and tendency at $0.05 < P \leq 0.20$. In Run 1, NC ($P = 0.014$) resulted in higher apparent organic matter disappearance (aOMD) while other treatments were not significantly different (NSD). However, NC ($P = 0.049$) and PV ($P = 0.029$) treatments also reduced apparent microbial biomass production (aMBP). In contrast, IB tended to increase aMBP ($P = 0.10$). For CH₄/OMD, IB, PV, and NC generated lower levels ($P < 0.05$) and SO₄ was NSD. In Run 2, EG resulted in less aOMD ($P < 0.001$), and OE ($P = 0.075$) and SN ($P = 0.189$) tended to result in less aOMD. SN ($P < 0.001$) and OE ($P < 0.001$) had less aMBP, and EG tended to have less aMBP ($P = 0.16$). For CH₄/OMD, EG tended ($P = 0.11$) to generate higher levels and all others NSD. STM mix showed an incredibly varied fermentation effect, with the IntelliBond mix resulting in a mitigation of negative impacts.

Session 6

Theatre 2

Factors affecting the in vitro kinetics of total gas and methane in dairy cows using Gas Endeavor System

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In ruminants, methane is a product of rumen fermentation, playing a significant role in greenhouse gas (GHG) emissions and dietary energy loss in cows. This study aimed to investigate the factors effecting total gas production (GP) and methane kinetics in different physiological stages of dairy cows (lactating: LC and dry: DC) using different buffer solutions (Menke: MS and Van Soest: VS). Gas Endeavor, an automatic gas flow measuring system (BPC Instruments, Lund, Sweden), was used to estimate GP and CH₄ in two runs. One gram of LC and DC diets was incubated with 150 mL of buffered rumen fluid (inoculum) using MS and VS buffer solutions and rumen fluids from LC and DC. Fermentation lasted for 24h at 39°C. Data were analyzed using linear fixed model with three factors: physiological stage, buffer solution and run. As expected, significant differences were observed in GP between physiological stages of cows (442 vs 288 mL/g of DM for LC and DC resp.; $P < 0.05$) and buffer solutions (345 vs 395 mL/g of DM for MS and VS resp.; $P < 0.05$). Similarly, the percentage of CH₄ in GP differed significantly for the physiological stages (15.3 vs 23.9 %; $P < 0.001$) and buffer solutions (21.0 vs 18.1 %; $P < 0.05$). The kinetics of GP and CH₄ exhibited their maximum rates between 6-7 hours of incubation (42.9 and 10.9 mL/g of DM resp.) Similarly, the kinetics of GP was significantly ($P < 0.005$) higher (17.61 mL/g of DM/hour) than that of methane (3.01 mL/g of DM/hour) considering both physiological stage and buffer solutions. In conclusion, physiological stage of cows and use of different buffer solutions were important factors in GP and CH₄ production. Furthermore, Gas Endeavor proves to be an innovative and suitable system for assessing both total gas and methane kinetics in real-time during in vitro rumen fermentation, that would be useful to evaluate nutritional strategies aimed to mitigate GHG emissions in ruminants. Funded by the project: PNRR, "National Research Centre for Agricultural Technologies – Agritech" – CN2, Missione 4, Componente 2, Investimento 1.4, CUP C93C22002790001

Impact of low-density polyethylene microplastic on ruminal degradability of feeds

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The primary plastic utilized in farms is low-density polyethylene (LDPE), often identified as a foreign object in the rumen of slaughtered cattle and in the form of microplastic (MPs, <5mm) both in ruminant feeds and their body tissues. Despite its widespread presence in farms, there is a notable absence of studies examining its potential impact on the ruminal degradability of feeds. This research endeavored to examine the influence of LDPE as MPs, on the in vitro rumen degradability of feeds. In 3 farms, 3 most used feeds were collected: mixed hay (Hy), corn silage (Cs), and concentrate (Cn). Feeds, dried at 60°C for 24h and ground (1mm sieve), were prepared in bags containing 0.5g on dry weight of feed, and incubated in a rumen fluid derived from Piemontese bulls. Three runs were performed. Four jars were added with 4 levels of LDPE (0, 0.5, 0.9, and 1.4% on rumen-buffer solution). Each jar contained 3 bags for each feed and each farm, for a total of 27 bags. They were incubated for 48h in the Ankom DaisyII with 400mL of filtered rumen fluid, and 1600mL of buffer solutions for jar. Results showed that LDPE at all levels did not affect feed degradability. Ruminal degradabilities (%), for 0 to 1.8% of LDPE, were: 47.6±8.92, 47.7±8.96, 49.2±8.72, and 48.5±8.76% in Hy; 59.1±3.97, 59.1±5.13, 59.1±4.86, and 59.7±4.24% in Cs; 69.2±5.80, 70.0±4.38, 71.6±5.28, and 71.6±6.35% in Cn. These results provide valuable insights into the resilience of the rumen degradation process in the face of exposure to LDPE. Future studies are needed to investigate the effects of other MP polymers to provide a deep understanding of the impacts of MPs pollution on ruminant nutrition.

Session 6

Theatre 4

Rumen contents from slaughtered cattle and sheep to study in vitro degradation of selected feeds

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Rumen degradation studies are needed to evaluate various feeds before their use to formulate nutritionally balanced diets for ruminant animals. However, the use of surgically modified animals for this purpose is highly regulated, laborious, and undesirable. Thus, suitability of rumen fluid (RF) from freshly slaughtered cattle (RFC) or sheep (RFS) was tested as a non-invasive alternative to estimate in vitro degradation of 12 feeds representing 6 concentrates and 6 grasses. Representative samples of concentrates and grasses were collected, dried and ground through a 1 mm sieve before their separate incubations with buffered RFC or RFS. Factorial studies were used to anaerobically incubate about 0.4g of each feed separately with 40 ml of either RFC or RFS for various times from 0 to 72 hours at 39°C. The undegraded residues were washed and dried to calculate degradation of each feed at each time before deriving constants and effective degradability (ED) by using a curve fitting model. The ED of dry matter (EDDM) or crude protein (ECPD) for RFS were comparable to those for RFC. While the degradability values differed ($P < 0.05$) depending on the feed and RF types, the values for forages and feeds compared well with those for fistulated animals. The feed ranking by degradability estimates was remarkably similar for RFS and RFC. Strong correlations were found between the two inocula ($R_2 = 0.86$ for EDDM and 0.74 for ECPD). The ECPD values for feeds in this study using RFS or RFC matched well with the reported literature values as well. This study showed that RF obtained from freshly slaughtered sheep or cattle could equally be used to replace RF obtained from fistulated animals for the in vitro incubation of feeds to estimate degradation of ruminant feeds.

Reducing lactating cows dietary sodium to reduce environmental cost of dairy farms

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This study aimed to examine the effect of reducing dietary sodium by reducing dietary NaHCO₃ or NaCl on lactating cow's intake, yield, and well-being. Excess dietary Na increases excretion in manure and washing water from the milking parlor. The Na can reach water reservoirs or open land, impairing their quality and harming the environment. We hypothesize that dietary Na can be reduced from ~0.47% to 0.31% of diet DM while supplying sufficient Na for health and production. The literature review supports this hypothesis; however, because Na can be added to the feed as NaHCO₃ or NaCl, it was necessary to evaluate the effect of Na concentration and its origin. After two weeks adaptation to group and individual feeding regime, 42 multiparous mid-lactating cows were blocked to groups of 3 according to days from calving, body weight, and yield, and each triplet randomly assign to one of the following treatments: (1) CON (typical lactation diet with 0.46% Na with added NaHCO₃ and NaCl); (2) NHC (0.31% Na with added NaHCO₃); and (3) NC (0.31% Na with added NaCl) for 5 weeks while recording DM intake, body weight, milk and milk solids (fat, protein, lactose) yield, lying time, activity (pedometer), and ruminal pH. Week 5 was dedicated to urine and fecal samples to analyze digestibility and mineral balance. Data were analyzed using the mixed-model procedure with treatment, day, and treatment*day as a fixed effect, cow as a random effect, and daily data as repeated measures modeled as AR(1). Regardless of reducing NaHCO₃ or NaCl, the model did not reveal a significant effect of dietary Na concentration on DM intake or energy-corrected milk yield that was 29.5, 31.2, 30.5, and 42.0, 41.1, and 41.3 kg/d for treatments 1, 2, and 3, respectively. However, higher dietary Na resulted in higher milk protein % (3.34 vs. 3.25) and a tendency (p = 0.08) to higher milk fat % (3.82 vs. 3.62) but not yield. Overall, reducing dietary to 0.31% will minimize the environmental cost of dairy farms without impairing production. Additional study is warranted before lowering dietary Na concentration below this.

Session 6

Theatre 6

Replacing hexane by 2-methyloxolane for defatting soybean meal does not impair fattening performance of beef cattle supplemented with methionine

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Chemical fat extraction for producing soybean meal (SBM) uses solvents like extraction-grade hexane (Hex). Concerns over health risks from n-hexane residues have motivated the exploration of alternatives like 2-methyloxolane (2-meOx), a bio-based solvent derived from agricultural by-products. However, differences in processing techniques between Hex and 2-meOx SBM may lead to varying nutritive values, potentially impacting animal performances. The aim of this study was to evaluate the performance of beef cattle fed grass silage diets with 2-meOx compared to hexane-extracted SBM, with or without supplementation of rumen-protected methionine (RPM). Thirty-six young Charolais bulls (248 ± 21.7 days) were studied during three consecutive 70d-feed efficiency tests until slaughter. Treatments resulted from a factorial design crossing two types of SBM (Hex vs 2-meOx) with 2 levels of RPM (supplemented or not). DM intake did not differ significantly among the four treatments for any of the three 70-day efficiency tests (P>0.05). Diets supplemented with RPM promoted higher average daily gain and feed conversion efficiency during the initial 70-day feed efficiency test, particularly in diets containing 2-meOx (SBM x RPM interaction; P≤0.02). Additionally, carcass weight, whole-body protein deposition, and nitrogen use efficiency increased with RPM supplementation, especially in diets containing 2-meOx (SBM x RPM interaction; P≤0.005). Our results highlight the potential of using 2-meOx as a bio-based solvent for SBM production in beef cattle diets but suggest a higher methionine deficit compared to Hex SBM.

2-methyloxolane can replace hexane for defatting soybean meal fed to cows without affecting dairy performance, rumen volatile fatty acid proportions and major fatty acid secretion in milk

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There are toxicity concerns regarding the use of hexane as a defatting solvent for soybean meal (SBM). 2-methyloxolane (2-meOx), a biobased solvent, could be an alternative but its use requires process adjustments. We aim to ensure that these adjustments do not affect the SBM feeding value for dairy cows. In a 4 × 4 treatments Latin-square experiment, 16 dairy cows received a diet supplemented with progressive substitution of hexane-defatted SBM by 2-meOx-defatted SBM: 100% hexane, 67% hexane + 33% 2-meOx (33% 2-meOx), 33% hexane + 67% 2meOx (67% 2-meOx), and 100% 2-meOx. The diet contained 16% SBM (on DM basis), 417 g NDF, 200 g starch, and 27 g crude fat per kg DM. We measured individual intake, dairy performance, rumen fluid volatile fatty acids (VFA) proportions, and milk fatty acid (FA) profile. Equivalence tests were used with equivalence margins defined as ± 5% of the estimated marginal mean of 100% hexane. Conclusions were drawn using the EFSA terminology. Dry matter intake and FA profile were equivalent among diets. FA intake was more likely equivalent than not. Rumen major VFA proportions were equivalent while those of minor VFA were likely lower, especially with the 67% 2meOx diet. Milk yield, fat concentration, fat yield, and FA profile were equivalent or more likely equivalent than not, except for C18:3 n-3 proportion which was likely reduced with 33% 2meOx and likely increased with 67% 2-meOx and 100% 2-meOx. Overall, 2-meOx can replace hexane for defatting SBM without affecting dairy performance, rumen major VFA proportions and major FA secretion in milk.

Session 6

Theatre 8

Effects of pre- and postpartum dietary management on performance in dairy cows

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The current study evaluated the effect of prepartum dietary energy content, postpartum protein content, and their interaction on performance and metabolic health in lactating dairy cows, in a 2×2 factorial design. Comparable studies were performed at two locations, including 60 and 48 Holstein cows in study 1 (the Netherlands) and study 2 (Canada), respectively. Prepartum treatments were a low energy diet (LED; ~85% NEL requirements) or a high energy diet (HED; ~130% of NEL requirements) fed for 21d prior to calving. Postpartum treatments were an average protein diet (APD; ~15.5% CP) or a high protein diet (HPD; ~18.5% CP) fed for 70 days post-calving. At implementation, the energy content of the prepartum diets were significantly lower in study 2 (9 and 6% for LED and HED, respectively). Relative to LED, feeding HED increased prepartum energy intake by 63 and 72% (P < 0.01) in studies 1 and 2, respectively. In study 2, feeding a HPD increased milk fat content in LED cows but decreased it in HED cows, resulting in a significant pre- by postpartum treatment interaction (P < 0.01). There were no other interactions. In study 1, feeding HED tended to increase milk fat content (4%; P = 0.06) and increased (P < 0.01) milk urea (8%) and body condition score (BCS; 3.2 vs. 2.9) in the first 9 weeks of lactation. In study 2, feeding HED increased (P ≤ 0.05) DMI (5%), milk (6%) and protein (5%) yields, and BCS (3.1 vs. 2.9); and tended to increase (P = 0.07) fat- and protein-corrected milk (6%) and body weight (4%), in the first 9 weeks of lactation. Feeding HPD increased milk urea in both studies (P < 0.01; 34 and 68% in studies 1 and 2, respectively). In conclusion, changes in prepartum dietary energy levels had the largest impact on early lactation performance.

Resilience to acute underfeeding in dairy ewes diverging in feed efficiency: rumen dimethyl acetal composition

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Certain lipids have been suggested as biomarkers of rumen function. This has encouraged the study of odd- and branched-chain fatty acids, but few data are available on dimethyl acetals (DMA), which are microbial lipids containing vinyl (alk-1-enyl) ether chains. Because rumen function may have a key role in feed efficiency (FE) and in the resilience to nutritional challenges, in this study we compared the pattern of variation in DMA in the rumen of dairy ewes phenotypically divergent for FE and subjected to a severe nutritional challenge (i.e., acute underfeeding). To estimate FE, intake and performance were recorded daily for 3 weeks in 40 Assaf sheep fed ad libitum a total mixed ration. Then, the highest (H-FE) and the lowest (L-FE) feed efficiency ewes (n=9/group) were fed only straw for 3 days. Before, during and 10 days after the challenge, ruminal fluid was collected using a stomach tube and DMA composition (as % of total DMA) was determined by GC-FID and GC-MS. A very dynamic pattern of variation was found: the challenge caused drastic reductions ($P < 0.001$) in the concentration of 7 DMA (e.g., DMA iso 15:0, 16:0, and t11- and c9-18:1), and increases in other 13 DMA (e.g., DMA iso 14:0, 15:0, anteiso 15:0 or 18:0). The DMA anteiso 13:0 could only be quantified during the challenge. All DMA recovered their initial value on day 10 after the challenge, and temporal changes in DMA did not differ between H-FE and L-FE. Overall, rumen DMA composition of ewes was very dynamic over time, but no relationship with FE was detected. Acknowledgments: PIE202340E068 (CSIC) and PID2020-113441RB-I00 (MCIN/AEI/Spain).

How does lamb feeding management during early life and high-cereal finishing affect the rumen trans fatty acid profile at weaning and the end of the fattening period?

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The aim of this work was to evaluate the effect of the pre-weaning feeding management on the rumen fatty acid (FA) profile of lambs finished with a high-cereal diet. After birth, 32 lambs and their dams were divided into 4 groups: lambs without their mothers and fed with milk replacer (M); lambs and ewes together in the pasture (P); dehydrated lucerne pellets (F); or concentrate for lactating ewes (C). Hay was provided in C and F groups. At weaning, lambs were housed in pairs (according to initial diet) and received a high-cereal concentrate and straw (ca 10%) until slaughter. Freeze-dried rumen contents, collected at weaning and slaughter, were directly transesterified and FA methyl esters were analyzed by gas chromatography. Data of C18 FA, expressed as % of total C18 FA, were analyzed using SAS, considering the effects of pre-weaning diet (D), phase (P, weaning vs end of fattening) and interaction DxP. Regardless of pre-weaning feeding management, the total biohydrogenation intermediates ($P=0.009$) and 18:1t10 ($P<0.001$) increased during fattening period. At weaning, M lambs had lower 18:1t11 proportions than other lamb groups (DxP, $P=0.018$), while at slaughter the 18:1t11 did not differ among groups. The 18:1t10/18:1t11 ratio was lower in P lambs and higher in M lambs than in other lambs at weaning, and until slaughter increased in C, F and P lambs and remained unchanged in M lambs (DxP, $P=0.043$). These results are useful to understand how pre-weaning feeding management influence the occurrence of t10 shift in lambs finished with high-cereal diets. Funding: Portuguese Foundation for Science and Technology (FCT) under projects UIDB/00276/2020 (CIISA), LA/P/0059/2020 (AL4AnimalS), UIDB/05183/2020 (MED), LA/P/0121/2020 (CHANGE), and PhD grant awarded to LF (2020.04456.BD).

Micronutrient Supply, Developmental Programming, and Strategic Supplementation

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Developmental programming is the concept that various stressors, including compromised nutrient supply, during developmental can result in both short and long-term changes in offspring. Large animal models of developmental programming demonstrate that perturbed maternal nutrition; including global under and over-nutrition, macronutrients, and micronutrient supply, including amino acids, vitamins, and trace elements can alter gestational development and offspring outcomes. Data indicate that strategic supplementation of micronutrients (Met, Arg, Se, folate, vitamin B12, choline, cobalt, sulfur and others) can alter placental function and fetal nutrient supply. Specifically in the offspring, multiple visceral tissues, metabolism, growth, and reproduction are impacted by strategic supplementation and/or compromised nutrition. Data demonstrate that compromised maternal nutrition and strategic supplementation alters transcript abundance, metabolomic profiles, and multiple metabolic pathways. The mechanisms underpinning developmental programming are driven by epigenetic events, which depend on one-carbon metabolism and specific micronutrient supply. The concept of developmental programming is strongly supported by data from ruminant animal models. Compromised maternal nutrition is a stressor driving programming events. Genomic and metabolomic changes in offspring are precipitated by altered maternal nutrition and during critical windows of development. The underlying mechanisms of developmental programming involve epigenetic events. Evidence suggests that strategic supplementation of micronutrients potentially mitigates negative aspects of developmental programming. Future research needs include; enhanced understanding of the mechanisms, practical relevance to production systems, impacts on animal health, and interrelationships with the host microbiome. Strategic micronutrient supplementation and precision nutrition approaches to fostering positive and mitigating negative aspects of developmental programming should improve efficiencies and sustainability in ruminant livestock production systems.

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Theatre 1

How can the seasonal climate changes influence horse's health and welfare in the extensive farming system?

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Keeping horses in marginal areas may trigger a transition towards more sustainable animal husbandry practices, resulting in favorable outcomes for the environment and animal welfare. The present study aims to describe how welfare and health condition of horses kept in marginal areas can be influenced by environmental and climatic conditions. A total of 90 horses (average age: 13.1±0.27 years) were recruited from seven stables in Italian marginal areas. The AWIN welfare assessment protocol was applied by two trained observers to monitor horses' welfare in summer and winter. The proportion of each score of animal-based indicators were calculated. During the winter, the percentage of horses with a suboptimal body condition score was slightly higher (62.9 %) compared to the summer (59.3 %). Altered hair coat condition (17.2 %) and cough presence (1.1%) were higher in the winter with, respectively, 9.9 % and 0 % in the summer. However, alopecia (19.7%), skin lesions (10.9%) and swollen joints (11.7%) were higher in the summer, while in winter they were 4.5 %, 4.5% and 4.8% respectively. Human-horse relationship was similar in summer and winter with over 80% of horses showing positive signs when approached by humans. The results suggest that different climatic conditions can affect the health of horses kept in marginal areas, thus affecting their welfare. Further data collection should aim to enlarge the time frame of monitoring, allowing a more comprehensive evaluation of the impact of different climate conditions on horse welfare.

Omic evaluation of resilience and adaptation to the combination of heat and exercise stressors in horse athletes

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Endurance race is one of the most challenging disciplines for horses which can induce metabolic imbalance especially if combined with sub-optimal temperatures. Incremental field standardized exercise tests (fSETs) are valuable systems for horse training and fitness evaluation. This research aimed to address molecular features of resilience and adaptation to a combination of heat and exercise-induced stresses in horses through the NGS approach. Six Arabian horses, stabled and homogeneously trained at the Italia Endurance Stable & Academy (Perugia, Italy), were monitored during fSETs in summer and winter, collecting blood samples before and after each fSET. Total RNA was extracted from serum and sequenced with Illumina® technology. Lactatemia and hematocrit levels significantly increased in summer vs. winter. From NGS data, a set of differentially expressed small RNAs were obtained and a protein-protein interaction network for targets of micro RNAs (miRNAs) was built. On this, a gene ontology (GO) enrichment analysis was carried out highlighting, in summer vs. winter, enriched terms related to: innate immune response, protein kinase activity and DNA methylation (for targets of up-regulated miRNAs); cellular response to cytokine stimulus, NF-kappa B signaling and signal transduction (for targets of down-regulated miRNAs). These preliminary results should be considered in outdoor sports, in animals as in humans, since the horse athlete is an optimal model to study exercise-related physical disorders sharing, with human athletes, poor performance syndromes, overtraining and potentially lethal consequences if the physical effort is associated with thermal stress.

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Theatre 3

Agonistic interactions at straw racks in group-housed horses

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As group housing of horses can be associated with an increased injury risk especially at limited resources, the aim of this study was to allow first scientifically based recommendations regarding positioning and design of straw racks. In a HIT active stable with 50 to 62 horses kept in one group, occupancy, feeding distance, prevalence of agonistic interactions (ais) as well as the percentage of feeding disturbances at two different straw racks were analyzed. Equine behavior was recorded for 6h/day on 15 summer and 15 winter days. Using generalized linear mixed models, considering the fixed effects rack, fresh straw supply and the interaction of observation day and time period delivered the best prediction for the number of feeding events/h and the prevalence of ais/h. Here, both racks differed significantly ($p < 0.05$) with twice as many feeding events/h, but threefold more ais/h respectively twice as many ais/horse at the larger rack, where conspecifics fed side by side. 65% of the harmless and 92% of the potentially harmful ais led to feeding disturbances. The latter made up 25% of all ais in total with no significant differences between both racks. Also feeding distances did not differ when quantified as number of free openings between two conspecifics (2.2 +/- 1.3) and we recommend an animal:rack opening ratio of at least 1:3. However, horses preferred to feed simultaneously with the rack edge in-between them at the smaller rack 1, so that regarding the head region, the actual animal distances were smaller there. Besides, the investigation of further straw racks and farms is essential to allow general statements, especially as our study proves many influencing factors.

Milking procedure and frequency: what influence on mares and foals behaviour?

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The aim of this study was to assess the impact of the 2hrs deprivation of suckling in foals (i.e. equipped with a muzzle) and of mares' milking frequency on the behaviour of both partners. We studied 23 multiparous mares divided in 3 groups: 8 non-milked mares as control (C); 8 mares milked once after 2 hrs (1M); 7 mares milked twice, once per hr (2M). Mares were milked at week 5 of lactation. Mares and foals were observed at pasture at weeks 4 and 6 using classical ethological methods to assess the quality of mother-young relationships (mare-foal distances, time with the dam as closest neighbour and frequency of suckling). They were observed during the milking procedure: after the muzzle's fitting and during the 2hrs of waiting, during milking and after the muzzle's removal. Before milking, we scored agitation in mares (agonistic behaviours), as well as frustration in muzzled foals (suckling attempts). Both were probably due, out of the situation (preventing from suckling/nursing), to the spatial and forage limitations within the waiting zone. Right after the muzzle's removal, almost all foals suckled without maternal opposition, confirming the procedure-induced frustration. However, no deleterious effects were scored in 1M and 2M groups when observed at pasture at week 6 (e.g. no shorter mare-foal distances or stronger mutual social preferences compared to the C group and week 4). Foals appeared to easily cope with the procedure probably as they were allowed to stay all along with their mothers. To conclude, the use of a muzzle appears to be a practice to promote in milk production industry. Nevertheless, further research is needed to determine the consequences of the repeated use of the muzzle associated with deprivation of suckling.

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Effect of antioxidant supplementation on faecal microbiota of horses at different exercise levels

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Diets rich in cereals can alter the horse digestive system, microbiota, fermentation patterns, and cause colic or laminitis. Although vitamin E (E) and selenium (Se) are antioxidants, Se improves fermentation in the rumen. Thus, our objective was to evaluate the effect of supplementation with Se (Se yeast) and E (α -tocopheryl) on the fecal microbiota of horses in moderate activity. Horses (n=24; 450 kg, 5-15 years) that had not been exercised the month prior to the study were used, housed, randomly assigned to 4 treatments in a factorial arrangement (2 Se \times 2 E, levels; LSe, 0.1; HSe, 0.3 mg Se/kg DM; LE, 1.6; HE, 2 IU E /kg BW; NRC, 2007). The treatments were: LSeLE, HSeLE, LSeHE, HSeHE. Since the daily basal ration was poor in Se, <2 μ g, and E, 14.4 IU/kg DM, supplementation (for 63 days) completely provided both nutrients. Feces were taken before treatments (1 w), after 4 weeks of supplementation and after 4 weeks of moderate exercise. Metagenomic analysis of the microbiota used the V4 hypervariable region of the 16S rRNA (Caporasso et al., 2011; Gaona et al., 2019). Purified rRNA amplicons were pooled and sequenced (Illumina MiSeq platform; Yale Genome Analysis Center, CT, USA). Sequences were analyzed with QIIME 2 2018.6 (Bolyen et al., 2018); a quality protocol was applied (Callahan et al., 2016). Taxonomy was assigned to ASVs (Rognes et al., 2016) against the SILVA database (132-99% OTU release, region 515-806, taxonomy L7) with a percentage identity of 0.95. The feature table was reduced to 32,184 ASVs per sample. Alignment was performed (Katoh et al. (2002). Alpha diversity was calculated (Shannon, Simpson and Observed otus; complement qiime alpha diversity). Phylogenetic β -diversity analysis was measured using weighted unifrac (Lozupone et al., 2007) and visualized (principal coordinate analysis (PCoA); complement qiime core-metrics-phylogenetic diversity at 32,184 ASV per sample). Preliminary results showed that microbiota α -diversity was affected by Se and exercise. Firmicutes was the main phylum in all treatments and weeks. Conclusion: the use of Se in the diet as well as physical activity modifies the fecal microbiota of horses.

A possible mathematical approach to predict forage digestibility from faecal particle size distribution in horses

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The relationship between forages' total tract apparent dry matter digestibility (ttDMD) and faecal particle size (FPS) was studied to develop a tool to predict forage digestibility in horses. The FPS and ttDMD of 6 horses, fed 6 different forages (4 cuts of fescue, perennial ryegrass/clover mix, and straw) were studied by a 6×6 Latin square design. Each period lasted 16 days (14 days of adaptation). Forage chemical composition was assessed by NIRs. Faecal and forage acid insoluble ash content were used to estimate ttDMD. The FPS was determined by wet sieving using a set of 6 sieves (from S1 to S6) of different mesh sizes (6.50, 4.60, 2.36, 1.18, 0.6, 0.15 mm). Horse and forage ttDMD's effects on FPS distribution by sieves were studied by GLM. Linear regressions and correlations between faecal residues on each sieve and the sieves' mesh were exploited to generate a mathematical tool to predict digestibility from FPS distribution. The FPS were unaffected by horse. Higher ttDMD were associated with larger FPS, with perennial ryegrass/clover and straw (ttDMD 74.44 and 44.57 %) leading to smaller and larger faecal particles. Strong correlations were found between ttDMD and S1 and S2 ($r = 0.639$ and $r = 0.668$; $P \leq 0.001$). The angular coefficient of the linear function between faecal residues on each sieve seems a promising tool for forage digestibility prediction from FPS ($r^2 = 0.041$). Thus, the determination of faecal residues of sieves of different mesh size can be used as an indirect tool to rank forage digestibility.

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Effect of phytochemicals on cyathostomins ecological interactions

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The emergence of cyathostomin isolates resistant to anthelmintics requires alternative control strategies, such as bioactive forages and their secondary metabolites. Their use in the field raises however questions about their effect on parasite ecological interactions. Here, we quantified the in vitro activity of two plant-derived extracts i.e. carvacrol and cinnamaldehyde, and established how increasing concentrations affected larval cyathostomin community structure in vitro using metabarcoding. *Cyathostomum pateratum*, *Cylicocyclus ashworthi*, and *Cylicocyclus nassatus* were the most abundant species in the absence of any treatment. Alpha diversity remained constant whatever ivermectin concentrations surpassing IC90, while species richness decreased significantly with pyrantel and plant extracts. However, absolute abundances of *C. nassatus* and *C. ashworthi* remained high at the highest concentrations of carvacrol, cinnamaldehyde and pyrantel, suggesting a lower sensitivity of these species. These results indicate that carvacrol and cinnamaldehyde selectively reduced the development of cyathostomin species, without infecting *C. pateratum*, *C. nassatus* and *C. ashworthi*. However, unlike the other two species, the chemical drugs were active on *C. pateratum*. While these phytochemicals display in vitro efficacy, they may reshape cyathostomin communities in the field.

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Possibilities and prerequisites for digital-technical systems in horse husbandry

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Digitalisation and mechanisation promise far-reaching changes on animal farms. Although horses are the predominant livestock in many German metropolitan regions and most of the work processes associated with horse husbandry are carried out manually, there has been little research into digitalisation and mechanisation on horse farms. This study therefore investigated which digital-technical applications (e.g. for manure removal, feeding, communication, health and safety monitoring) currently exist on horse farms and which prerequisites are required for establishing the systems. For this, an online survey was conducted with 451 stakeholders on horse farms in Germany via Google Forms. Results show that (in % of total participants) cameras for safety (30.8 %) and health monitoring of horses (22.6 %) as well as customer communication systems (24.8 %) and farm management software (13.7 %) were most frequently mentioned as existing. Other technologies, such as digitally controlled automated feeding-systems for roughage (7.3 %) and concentrate feed (9.3 %), automation of manure removal (3.3 %) and digital monitoring of stable air parameters (2.2 %) were stated by less than 10 % of the participants. The type of housing (group or individual housing), a stable internet connection and the willingness of farm managers to invest have a particular influence on the use of digitally supported systems. Digital technology was used more frequently on farms with a reliable internet connection and a division of horse husbandry into functional areas. The benefits of the systems for all those involved (horses as well as farm managers and employees) should be clearly communicated and the appropriate use should be reviewed on an individual farm basis.

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Air quality welfare factors in horse husbandry - Dusk pollution and digital technology

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Background: The digital measurement of stable air parameters is crucial in modern horse husbandry. By monitoring parameters such as temperature, humidity, ammonia content and dust concentration, the risk of respiratory diseases for horses linked to stable conditions can be reduced. These technologies offer the opportunity to quickly recognize deviations from ideal conditions and react accordingly. Material and methods: We focused on analyzing particulate matter concentrations in three specific areas: inside the stable, in the outside air and in the immediate vicinity of the horses' nostrils. Three simultaneously measuring particulate matter sensors were used, with one of the sensors being attached directly to the horse's holster. Air velocity, humidity and temperature were digitally recorded. Particulate matter emissions, such as oats and watered milled hay cobs, roughage, including loose hay and hay offered in hay nets, were investigated. The particulate matter effects of straw and steamed straw were also investigated. Results: The highest concentrations of particulate matter occurred when feeding oats. Milled and moistened hay cobs showed low particulate matter levels. A significant level of particulate matter was recorded when hay was used in hay nets. A remarkable 75% reduction in particulate matter was at steamed hay. Conclusion: These results underline the importance of feed preparation methods in minimizing exposure to particulate matter in horse husbandry and provide valuable insights for the development of strategies to improve air quality in stables.

Evaluation of shelter design on equine rhythmicity

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Nowadays, the assessment of equine well-being commonly involves observing equine behavior, body language, acoustic signals, and physiological indicators. Another approach for assessing an animal's welfare involves investigating rhythmicity. Rhythmicity, which represents the synchronization of an organism with its environment over a 24-hour period, is less susceptible to temporary factors. Consequently, the analysis of rhythmic patterns may serve as an objective indicator of animal welfare. One can calculate a rhythmicity index called DFC (Degree of Functional Coupling) from temporal data, such as daily locomotor activity. The present study aims to assess how different intrinsic motivations to rest affect horses' rhythmicity. Twenty horses divided into four groups and housed in a paddock trail group housing system were studied during four winter months. All horses could move freely and had free access to water and the littered resting area (shelter). Horses were fed with an automatic feed dispenser, which gave them access to hay three times per day for two hours each. Further, all horses were able to access straw which was provided in the shelters. We investigated the effect of four different shelter design treatments on rhythmicity: closed shelter (wood shelter with three exits), large entrance (single large entrance along one long side of the wood shelter), sand (wooden shelter with three exits and addition of sand in front of the shelter), and large entrance + sand (single large entrance along one long side of the wooden shelter and addition of sand). Each group of animals experienced each of the treatments for four weeks (2 weeks of habituation followed by 2 weeks of data collection) following a Latin square design. All horses were equipped with an accelerometer during the data collection period. The DFC and lying behaviour were computed from this temporal data. Statistical analysis has been performed with a linear mixed effect model. We observed changes in DFC within horses and across treatments. The whole results will be presented during the conference.

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Time-controlled hay racks in group-housed horses: What ratio of horses to feeding places is appropriate?

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Automated feeding systems offer numerous advantages. Nevertheless, practical experience shows that individual distances at time-controlled hay rack for horses are frequently undercut and aggression can therefore increase. In the present study, the minimum animal-to-feeding-place ratio (AFR) was investigated at time-controlled hay racks, which would ensure sufficient individual distance between the horses. Using ethological and physiological parameters of 28 horses in a group housing system, four treatments were assessed on two observation days each: an AFR of 1:1.2 (20% more feed-throughs than horses), 1:2 (twice as many feed-throughs as horses), 1:3 (three times as many feed-throughs as horses) and C (individual feeding in the grooming area with familiar conspecifics). The order of treatments was randomized, with one treatment per day and at least one day of wash-out phase between treatments, thus extended over a period of 3 weeks (C, 1:3, 1:1.2, C, 1:1.2, 1:3, 1:2, 1:2). Horses were used to daily fluctuation of the group composition, as horses were taken out of the group for riding by their owners every day and at different times, even during feeding. A preliminary test was conducted to habituate the horses. The horses showed less aggressive behavior when there were three times as many openings as horses at the time-controlled hay racks compared to 1:1.2 and 1:2 (lineal mixed model: $F(3,4) = 7.411$; adjusted $R^2 = 0.733$; $p(\text{AFR}_{1:2}) = 0.06$, $p(\text{AFR}_{1:3}) = 0.02$, $p(\text{AFR}_C) = 0.01$). Salivary cortisol levels during feeding decreased less with low AFR ($p(\text{AFR}_{\text{metric}}) = 0.02$). The results indicate that at least three times as many feeding places as horses must be available for the investigated time-controlled hay racks so that the horses can maintain their individual distance. Further studies are required to validate the results.

Thermographic Images as a Diagnostic Tool for Equine Headshaking Syndrome

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Equine headshaking syndrome in horses consists of spontaneous, uncontrollable movements of the horse's head and neck in the absence of external stimulus. Stress-related causes as well as inflammations, amongst others of the N. trigeminus have been discussed. At the same time, thermographic imaging is being increasingly used in veterinary and animal sciences with regard to the detection of stress, negative emotional states and inflammations. Therefore, we analysed whether thermographic imaging could serve as a diagnostic tool for equine headshaking syndrome, following the hypothesis that causes are stress-related or linked to inflammations and therewith detectable via thermographic imaging. 223 thermographic images were gathered of 17 horses experiencing idiopathic headshaking and 13 control horses during standardised riding tests. Statistical analysis was performed to examine the variance in eye temperature pre- and post-riding. For this analysis, a mixed model was created for thermographic images, which included the difference in eye temperature before and after riding. The model included the fixed effects breed (horse, pony), sex, presence of headshaking as well as the rider (owner, external) and the covariable environmental temperature. Descriptive statistics revealed that the inner eye temperature increased by 0.13 degrees after riding. Out of 135 images captured for the left eye, a higher eye temperature was observed after riding, while 88 images displayed a lower eye temperature post-riding. No significant differences were observed in thermographic images between headshakers and control horses. In order to be able to use thermographic images, interference factors must be taken into account. These include standardised distances and recording angles, lighting conditions, weather influences and other environmental factors. Standardisation proved to be difficult in this study due to the long observation period and thus different climatic conditions. The use of thermography proved to be impractical and showed no effect in the experiment.

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Theatre 13

Anatomical bridles, do they really alter and reduce pressure distribution on the horse's head?

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Various bridle designs, like the "anatomical bridle", are commercially available. Many manufacturers claim reduced poll pressure and improved equine performance, without convincing scientific evidence. Ill-fitting tack such as over tight nose bands and head pieces that cause pressure points can influence unwanted behaviours and physiological detriment such as head shaking and restricted blood flow to key facial structures. This study aimed to investigate the impact of two bridle designs, a standard Cavesson and Anatomical Bridle, on headpiece (HP) and noseband (NB) pressures. Multiple long reining trials were conducted at University Centre Myerscough using Tekscan pressure sensing technology to compare HP and NB pressures between bridles and different exercises. Higher peak pressures ($P < 0.05$) were recorded with the cavesson across the HP compared to the NB, with the anatomical bridle demonstrating the opposite. Peak pressures under the HP and NB were highest when working on a half-circle compared to a straight line ($P < 0.05$); lowest peak pressures were recorded under the HP on a full circle, whilst lowest NB pressures were recorded a straight line ($P < 0.05$). Regarding different paces, highest peak pressures were recorded in walk under the NB ($P < 0.05$). These results suggest that there may be an overall pressure exerted by any bridle across the facial structures of the horse and dependent on bridle design, their pressure distribution is altered accordingly. Further study is needed with a larger sample of horses, but these results indicate that horses may require individualised assessments for bridle fit and design.

The possibilities of using genomic information in the selection of horses for meat production

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Horsemeat, valued for its nutritional superiority over beef, faces cultural and ethical barriers to acceptance. Despite its benefits, genomic research on horsemeat production remains sparse, with no QTLs specifically identified. Our study aims to bridge this gap by prioritizing SNP markers on the GGP Equine chip associated with meat production traits, focusing on orthologous genes from cattle. We identified 30 genes within QTLs for dressed carcass muscle weight and dressed carcass muscle-to-bone ratio in cattle, leading to the identification of 22 horse orthologous genes, all located on chromosome 3, in a syntenic block rich with growth-related QTLs in horses, such as body weight, size, and chest circumference. Considering variant consequences, genomic evolutionary rate profiling scores, and the potential location of regulatory elements, we identified several SNP markers with potential impacts on protein function and gene transcription, including within genes like ADGRA3, LGI2, PCDH7, PI4K2B, PPARGC1A, SEPSECS, and SLIT2. These genes are implicated in key biological processes such as fatty acid synthesis, metabolic pathways, mineral absorption and metabolism, and fetal growth, underlining their potential in enhancing meat production traits. The objective of our research is to design the structure of a database, incorporate various data into the database, and develop technical applications that will enable the systematic improvement of meat production traits, like methodologies applied in cattle research. The data and applications for the use of the results will be organized within the Digitization of Livestock Databases (DigŽiv) project, funded by the Recovery and Resilience Plan and the Ministry of Agriculture, Forestry and Food, to support routine selection work for both breeders and professional services. Additionally, the principles of this approach will be applicable to other traits and other species. Validation through real data and comprehensive genetic analyses, including GWAS and Genomic BLUP, are essential for incorporating these findings.

Session 7

Poster 15

Influence of head hair whorls on behavioral measurements assessed with infrared thermography and behavioral surveys

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Pura Raza Español horse (PRE) is a worldwide breed, with a calm and noble character, used for Dressage performance. During the sport events, horses are exposed to several environmental stimuli that could affect their sport performance due to the stress developed. Hair whorls (HW) are described as changes in the hair pattern and the association between their location and equine temperament has been reported. The aim of this study was to assess the influence of HW on the equine behavior measured with eye temperature (ET) and a behavioural survey. ET was assessed with infrared thermography and collected from 98 PRE while performing a dressage exercise. A total of 18 horses showed an average of 1.38 HW in forehead, 59 showed an average of 1.22 between the eyes and 37 PRE showed an average of 1.15 HW below the eyes. During the dressage exercise, 3 ET samples per animal were obtained one hour before (BE), just after (JAE) and one hour after (AE) the exercise. To evaluate effort and recovery levels, a previous ET variation (PETI=JAE-BE) and a later ET variation (LETI=JAE-AE) were calculated. The influence of HW on the effort (PETI) and recovery (LETI) stress suffered by the PRE was studied with a multiple trait unifactorial GLM test, and a post-hoc Tukey's Test. Results showed significant differences in PETI for presence (-0.39C°) or absence (1.07 C°) of forehead HW. The correlation between effort stress and the number of above HW was -0.35. Thus, horses with HW in forehead showed a lower effort stress. Two behavioral survey variables, learning and cooperation, showed a significant Spearman rank correlation (0.21 and 0.22, respectively) with the HW between the eyes. These results are preliminary and require further research, but we believe they will be of great interest to breeders, especially those who participate in competitions.

Inter- and transgenerational impacts of in ovo nutriepigenetic intervention on chicken male gonadal transcriptome

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This study aims to explore for the first time the intergenerational and transgenerational impacts of in ovo injection of bioactive substances on gene expression in male gonads of green-legged partridge-like chickens over three generations. Synbiotic PoultryStar® (Biomim) and epigenetic factor choline were injected in ovo on the 12th day of egg incubation. Three groups were established in generation F1: 1/control (C, 0.9% physiological saline); 2/ synbiotic (S, 2mg/embryo); 3/ synbiotic combined with choline (SCH, 2mg/embryo of synbiotic and 0.25mg/embryo of choline). In the F2 and F3 generations, groups S and SCH were divided into two groups each: A/ single injected in F1; B/multiple injected in each successive generation. At 21 weeks post-hatching, cockerels in F2 and F3 were sacrificed and gonadal tissues were sampled. RNA was extracted for transcriptomic sequencing on Illumina Novaseq 6000 (PE 150). Repeated injections of synbiotic and choline influenced gene expression in gonads compared to the control and synbiotic alone in both F2 and F3 generations. Comparisons between analogous groups across generations showed enriched KEGG pathways particularly in downregulated pathways such as metabolic pathways, pathogenic infection pathways, and pathways associated with cancer, following repeated administration of synbiotic and choline. Protective regulatory mechanisms in gonads through those pathways can be suggested. A comprehensive understanding of the impact of the nutriepigenetic substances on gonadal tissues and their potential for transgenerational inheritance will be concluded based on ongoing analysis across all groups. This research was supported by the National Science Centre, Poland (grant no. 2020/37/B/NZ9/00497).

Session 8

Theatre 2

Early life profiling of weaning robustness using productive performance and gut microbiota composition
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Weaning is a critical moment of the pig productive cycle, and finding robustness-linked traits to improve robustness at weaning is crucial. With this objective, 218 piglets were weighted at birth (BWd0) and d25 (BWd25) and d42 of age (BWd42). Occurrence of diarrhoea was collected one week after weaning at d34. Faecal DNA microbiota was analysed by 16S sequencing from faecal swabs collected at d4, d25 and d34. Analysis of performance data using generalized linear model revealed the variables ($P < 0.05$) BWd0 associated with Average Daily Gain from birth to d25 (ADG0_25), and from d25 to d42 (ADG25_42). On gut microbiota data, PERMANOVA analyses of beta-diversity revealed associations between microbiota composition and host variables at the three time points. BWd0 was significantly associated with the microbiota of piglets at d4. Microbiota at d25 was associated with ADG0_25 and diarrhoea. Finally, microbiota at d34 was linked to diarrhoea and ADG25_42. In agreement, differential abundance analysis revealed groups of ASVs (Amplicon Sequence Variants) linked to several variables at each timepoint. At d4, 2 ASVs belonging to Clostridia class were positively linked to ADG0_25. At d25, 2 ASVs belonging to Clostridia class were negatively linked to diarrhoea. At d34, 118 ASVs were positively (47% belonging to Clostridia and Bacteroidia classes) and negatively (29% belonging to Clostridia class) associated to ADGd25_42. Overall, our study reveals potential early-life microbiota predictors linked to diarrhoea and ADG, which may confer to piglets a higher robustness to weaning.

Intestinal health status and dietary spray dried porcine plasma inclusion on faecal calprotectin and performance in weaned piglets

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Calprotectin (Cal) is a protein released by neutrophils in response to intestinal inflammation, which could be used as inflammation biomarker during post-weaning period. The aim of this study was to set the calprotectin levels in two Intestinal Health status (IH) and the impact of plasma inclusion on intestinal inflammation. Two farms were selected regarding their IH, one with a High-IH (HIH) and other with Low-IH (LIH), involving 294 and 380 piglets [(LD x LW) x Pt], respectively. Animals were distributed into 12 pens by BW and randomly assigned to a diet with 4% plasma (P; SDPP 80% CP) or control (C; plain diet) during the pre-starter phase (PS). Feed intake was monitored in the PS, while piglets' growth throughout the nursery period. At day 3 and 10 post-weaning, faeces were collected from the average BW piglet per pen and calprotectin was quantified using a Bühlmann fCAL turbo test. Results were analysed as a 2x2 factorial arrangement through an ANOVA test with R software. An interaction between diet and IH showed a higher BW and ADG, and lower FCR at the end of the PS for P-LIH compared to C-LIH piglets and C-HIH compared to P-LIH ($P < 0.05$). However, BW, ADG and FCR were not affected by P in the HHS farm ($P = 0.9$). No interaction was observed for Cal ($P > 0.1$). On days 3 and 10, higher Cal levels were observed in LIH than HIH ($P < 0.001$), and the plasma inclusion reduced Cal levels regardless the IH ($P \leq 0.05$). To conclude, the Cal levels suggest a lower intestinal inflammation and higher performance in HIH than LIH. In addition, plasma inclusion may improve intestinal health in weaned pigs.

Session 8

Theatre 4

In pursuit of a self-assembling protein decoy to counter Enterotoxigenic Escherichia coli F4+ strains causing piglet post-weaning diarrhea

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Post-weaning diarrhea (PWD) can cause severe losses in industrial pig farming. It is frequently caused by enterotoxigenic Escherichia coli (ETEC), expressing F4 or F18 fimbriae that bind to the intestinal microvilli of the piglets after weaning. Concerns about the emergence of antimicrobial resistance contamination have been raised globally due to the excessive use of antibiotics and zinc oxide to treat PWD. Therefore, alternative approaches to treat ETEC and preventing PWD are urgently needed. This work explores the use of self-assembling nanoparticles as decoys to counter intestinal adhesion of ETEC F4+ to microvilli. mi3, an icosahedral self-assembling protein, consisting of 60 monomers that form the mi3 scaffold, is bioengineered to display anti-ETEC F4 single domain antibody fusions at the N-terminus of each monomer. The expression of the recombinant protein was optimized and purification of protein by Size Exclusion Chromatography and validated by immunoblotting and Atomic Force Microscopy. In vitro tests showed that the anti-ETEC F4-mi3 nanoparticles bind ETEC bacteria and trigger its accumulation up to 5% of aggregation, leading to their incapability of binding intestinal microvilli. These promising results were confirmed by microscopy assays and through western blot analysis and will be further tested and optimized. Currently, also other nanobodies recognizing different F4 fimbrial epitopes are tested.

Oral alternatives for intramuscular iron administration in suckling pigs

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Piglets are routinely supplemented with intramuscular iron dextran injections to prevent anemia. However, this method is labor-intensive and not without risk. Our study compared the standard iron injection with various oral supplementation methods to evaluate their effects on hemoglobin (Hb) levels at weaning and growth performance during lactation and post-weaning. A total of 650 piglets from 46 litters was allocated to five treatments: standard intramuscular injection (IM), three iron-rich dry feeds provided ad libitum during the first two weeks after birth (O1: 10-12% Fe₂O₃, O2: 22.5% FeSO₄.H₂O, O3: 24% iron mixture) and a fifth treatment involving a blend of O3 with applesauce that was individually administered (O3 APPLE). The Hb levels at weaning of the piglets that received the O3 (6.7 ± 1.3 mmol/L) and O3 APPLE (7.7 ± 0.7 mmol/L) treatment did not differ significantly from the IM piglets (7.3 ± 0.9 mmol/L). The two other oral iron supplements resulted in significantly lower mean Hb levels with 4.8 ± 1.2 and 6.0 ± 1.3 mmol/L for O1 and O2, respectively. In the first period post-weaning (weeks 4-6), piglets of the O3 APPLE group had a higher growth rate (176 ± 69 g/d) compared to O1 (104 ± 89 g/d, p = 0.008) and O3 (122 ± 72 g/d, p = 0.04), but not significantly different from IM or O2. However, considering the overall lactation and nursery phases, growth was not affected by the iron treatments. In summary, the results indicate that orally administering iron can achieve an iron status comparable to that achieved by standard injection.

Session 8

Theatre 6

Effect of authorized level of Zn supplied in three different forms on the physiology and performance of piglets weaned at different age

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This study aims to assess the effect of three sources of zinc (Zn) and weaning age on piglets' performance and gut health status. At weaning, 96 pigs were arranged in a 2×3 factorial design including weaning age: Early (E, 21 days) or Normal (N, 26 days) and ~120 mg/kg in feed of the Zn from 3 sources: Zn sulphate (CO), Zn Glycinate (Gly) and potentiated form of Zinc Oxide (Hi). Piglets were weighed at weaning (d0) and weekly until d21. On d8 and d21, 48 piglets (one piglet/replicate) were slaughtered and jejunum tissue was collected for genes expression and morphology analysis. Data were analysed using a linear mixed and ANOVA model including diet, weaning age and their interaction as fixed factors and litter of origin as random factor. Weaning age affected the body weight through the entire study (P<0.001), resulting higher in N piglets. The interaction between diet and weaning age tended to affect the gain to feed ratio at d7-d21 (P= 0.07) which tended to be higher in EHi compared to the EGly (P= 0.09). Villus height and villus height to crypt depth ratio (VH:CD) tended to be affected by the weaning age at d8, resulting higher in the N group (P<0.10). GPX-2 expression was higher in NGly compared to NHi at d21 (P= 0.02). At d21, crypt depth was lower in EHi than ECO (P = 0.02); VH:CD was higher in EHi compared to ECO (P= 0.04). The administration of the potentiated form of ZnO had the same effects as Zn Glycinate, contributing to reduce the piglets' detrimental effect on weaning on gut homeostasis.

Uncovering the genetic background of porcine congenital splay leg syndrome in piglets

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Porcine congenital splay leg syndrome (PCS) is the most common congenital disorder in pigs and the leading cause of lameness in newborn piglets. Animals affected by PCS exhibit a splayed position of the hindlimbs, reducing their ability to stand and walk. Despite the transient nature of the disorder, mortality can reach up to 50% due to overlying and starvation. Although the condition has a significant impact on pig farming, the genetic background remains unknown. In this research, a genome-wide association study (GWAS) was conducted on 1224 Piétrain boars, genotyped on the GGP Porcine 50K medium density SNP chip. For these boars, data was collected on 12700 crossbred litters (Piétrain sire, hybrid dam) which were scored for the number of PCS piglets per litter. After quality control 43320 SNPs remained available for further analysis. Two strategies for GWAS were performed with the PLINK software. First, boars were divided into 'cases' (n=39) and 'controls' (n=650) based on their number of offspring and the prevalence of PCS. Second, a quantitative GWAS approach was used. The subsequent study revealed two significant regions on chromosomes SSC4 and SSC14 with signals reaching significance up to 10^{-10} . Moreover, a literature review on these loci identified strong candidate genes related to muscle physiology. These candidate genes are consistent with the prevailing notion that PCS correlates strongly with a delayed muscle development. Although additional research is necessary to validate these initial findings, this study provides a promising foundation for further exploration. In this manner, we have progressed another step towards elucidating the genetic underpinnings of PCS.

Session 8

Theatre 8

Interobserver reliability of different methods to evaluate sows' body condition

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Maximizing production and ensuring sustainability are two central goals of modern pig farming, and in order to achieve that is crucial to prioritize an adequate body condition of sows. It's crucial to routinely evaluate the body condition of sows to ensure a higher percentage of females are maintained in their optimal physical state. Different methods are available to measure sows' body condition and in the literature, there is no consensus of which method is the most reliable. The aim of this trial was to compare methods to assess sows' body condition between different observers. The methods used were the caliper, a mode A ultrasound (Lean-meter; Renco, Minneapolis, MN, USA) and visual body score (ranging from 1 – 5). For the measurements, 22 sows of different gestational ages were chosen and 8 observers performed the measurements in all the sows. The coefficient of variation was calculated for each method and individually for each sow. An average of the coefficient of variation for each method was then calculated. The difference between the accuracy of the method was analyzed by ANOVA and statistical significance was set at $p < 0.05$. There was no difference ($p > 0.05$) between the coefficients of variation of the caliper (CV = 7.7%) and the ultrasound mode A (CV = 5.3%) and both were lower ($p < 0.05$) from the visual body score (CV = 18.8%). Factors that may have influenced the results include observer bias and the level of observer training. Addressing the latter might require additional focus, particularly in subjective assessment methods. Objective methods, that are not interfered by, have a better precision to measure sows' body condition.

The secret of each sow: exploring the impact of sow and litter features on within-litter uniformity
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Variation in piglet body weight causes challenges in nowadays pig farming. Lower litter uniformity is associated with a higher occurrence of lightweight piglets and increased pre-weaning mortality. This study aimed to explore different metrics for quantifying within-litter uniformity, as well as to investigate associations between sow- and litter-specific factors and within-litter uniformity. We analyzed data from 610 litters of hybrid sows mated to Piétrain boars where 9,319 piglets were individually weighed at birth. Six different metrics were used to score within-litter uniformity: coefficient of variation (CV), standard deviation, average Z-score, range in weights, percentage of piglets < 0.8kg and < 1kg. Pearson correlations between these metrics were all positive and ranged from 0.22 to 0.83. The uniformities of consecutive parity litters are correlated ($R=0.27-0.43$), except between 4 and 5. Moreover, we found that CV of birth weight was associated with parity, number of pigs born alive, percentage of mummies, percentage of stillborns and service year. The positive association between CV and the proportion of mummies ($p=0.001$) and stillborns ($p<0.001$) suggests that fetal deaths or uterine environment changes linked to mummies may decrease litter uniformity. Additionally, 16% of the total variability in CV% can be attributed to differences between individual sows. Our results indicate that it might be feasible to quantify the uniformity of a litter by only recording the number of lightweight pigs and that within-litter variation can be linked to the features of each individual sow.

Effect of top-dressed arginine supplementation in different gestation periods and seasons on the colostrum composition and productive performance of sows and piglets
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This study aimed to evaluate the effect of supplementing 21.8 g/day of top-dressed arginine (Arg) at different gestation periods and seasons on the productive performance and colostrum of sows. A total of 320 sows were divided into 4 groups during 2 seasons (summer or winter): a control group (CO) fed a standard diet and 3 groups supplemented with 21.8 g/day Arg either during the first 35 days (EARLY35), or the last 45 days (LATE45), or during all gestation (COM). Colostrum was collected and analysed for composition, Igs concentration and metabolomic profile. Individual piglet body weight (BW) was recorded at birth, d6, d28 and 14 days post-weaning. Data were analysed using a GLM model including diet, season and parity as fixed factors. Interaction between diet and season was not significant. EARLY35 group had a lower percentage of stillborn and low-birth BW piglets than the other groups ($p<0.05$). COM group had a higher percentage of crushed piglets than CO group ($p<0.001$). LATE45 group had a higher percentage of high-birth BW piglets than the other groups ($p<0.001$). No difference in colostrum composition was observed, while Arg groups had less IgG than CO group ($p<0.001$). CO group had a higher concentration of colostral UDP-GlcNAc than Arg groups ($p<0.05$), which had reduced creatine and related metabolites. In conclusion, 21.8 g/day of on top-dressed Arg had a negative effect on the productive performance of the sows when given throughout gestation, while Arg supplementation during the first 35 days of gestation led to reduction in the percentage of stillborn and low-BW piglets.

Impact of replacing diet soybean with spirulina on blood metabolic profile of heavy pigs

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This study evaluated the feasibility of replacing dietary soybean (SBM) with spirulina (SP) on a set of blood metabolites in 88 gilts and barrows housed in 8 pens. Pigs were assigned to 4 diets (2 pens/diet): a commercial control diet based on SBM and cereals and 3 diets where SBM was replaced at 33, 66 and 100% (SP100) with SP. Blood samples were collected from all the pigs before the administration of SP (T0) and 145 days later, at the end of the trial (T1). Data were analysed i) keeping T0 and T1 separated according to a mixed model including sex and diet as fixed effect, and pen within diet as random effect, and ii) including in the model also the fixed effects of the blood sampling period and its interaction with diet, and the animal random effect. Diet did not affect blood metabolites both in T0 and in T1. When considering T0 and T1 together, the interaction period x diet was significant only for metabolites associated to hepatic overload and oxidative stress. Concentration of albumin, antioxidant rate and γ -glutamyl transferase increased regardless of treatment. Conversely, aspartate transaminase exhibited a lesser increase with the SP100 diet compared to C after the fattening period. Overall, the changes in blood metabolites were not related with the increase in SP content in the diet. Our results indicated that the dietary replacement of SBM with SP did not impact blood metabolic and inflammatory indicators, suggesting a good adaptation of pigs to the novel protein source.

Session 8

Theatre 12

Effect of soybean reduction and crude protein restriction on the performance of fattening pigs

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Imported soybean contributes to the environmental impact of the EU pig production. This study aimed to assess the impact of the reduction of soybean meal (SBM) and crude protein (CP) content on the performance of Italian heavy pigs. At housing, 1930 pigs with an average body weight (BW) of 36 kg were divided in: i. control group (CO), fed a standard diet; ii. treated group (TRT), fed a diet formulated to reduce SBM by 31%, 67% and 69% (replaced by sunflower and pea) in the 3 feeding phases (FP) and 1.2% CP in the last feeding phase. From 251 pigs, individual BW, Average Daily Gain (ADG), feed intake and Gain to Feed ratio (G:F) were recorded and faecal ammonia (NH₃) was determined from 20 pigs/group. Welfare indicators were monthly recorded. At slaughter (BW=180 kg), carcass and meat quality (L. lumborum muscle) were assessed. A linear mixed model (diet and sex: fixed factor; pen: random factor) was adopted. The TRT diet improve the overall ADG ($p=0.04$) and G:F ($p=0.01$), reduced FI ($p<0.0001$). The lesions score index at d102 was reduced ($p=0.03$) and promoted Methanobrevibacter (d94; $\text{padj.}=0.013$) and Clostridium sensu stricto (d181; $\text{padj.}=0.001$). Diet did not affect faecal NH₃, carcass composition and meat quality traits. In conclusion, replacing ~56% of SBM with more sustainable feed ingredients combined with a CP restriction is a promising strategy to formulate finishing diets with a reduced environmental impact.

Analysis of the effects of temporary outdoor access during the fattening phase on pig welfare, health and growth

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In the European Union, less than 1% of pigs are raised with access to outdoor areas. In this study, we analysed the effects of giving pigs temporary access to a pasture during the fattening phase on their welfare, health, and growth. We followed 300 pigs in 3 batches, reared in a conventional barn either without any outdoor access (O-) or with access to a pasture twice a week in the mornings, from D76 to D150 (O+). We measured at different timings their growth, stress level and immune competence and assessed body lesions. Pigs were weighed on D70, D133 and D150. We measured the level of salivary cortisol on D104 and D150. Monocytes, neutrophils and lymphocytes were counted from a blood sample on D135. On D150, we quantified IgA levels in saliva and scored body lesions. Linear mixed models were used to evaluate the effects of outdoor access and sex on these variables. According to preliminary data obtained on 2 batches, outdoor access had no impact on growth. On D104, O+ pigs exhibited lower salivary cortisol levels than O- pigs. On D135, we observed that both male and female O+ pigs displayed higher number of monocytes than O- pigs. In contrast, the neutrophil-to-lymphocyte ratio was higher in O+ females only. Finally at slaughter, the levels of salivary cortisol and IgA were significantly lower for O+ pigs, which also displayed fewer body lesions than O- pigs. Our results suggest that outdoor access had a positive impact on pig welfare, health and growth. Altogether, our study should provide new knowledge for a better understanding of the consequences of outdoor access on the pigs.

Session 8

Theatre 14

Effects of different olive cakes inclusion in the diet of the Bísaros pigs on the chemical composition and fatty acids profile of Biceps Femoris muscle

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Significant quantities of olive cake (OC) are generated by the olive oil industry, and their utilization in animal feed presents economic and environmental interest. This study aimed to evaluate the impact of different olive cakes (OC) in the diet of Bísaro pigs on the chemical composition and fatty acid profile of the biceps femoris muscle. Forty Bísaro pigs (castrated males and females; growing-finishing) were distributed across 5 treatments: Control, 10% inclusion of crude OC, Two-Phases OC, Exhausted OC, and Exhausted OC +1% olive oil (CD, COC, TPOC, EOC, and EOCCO, respectively). The diets significantly influenced the moisture content (%) of this muscle ($P < 0.05$), with the TPOC group presenting the highest moisture content (74.20) and the CD group the lowest (70.41). Protein content (%) was also significantly altered ($P = 0.0002$), with the CD group (20.91) being statistically different from the groups with OC incorporation (mean of 22.61). The heme pigments content (mg/g) was also significantly affected ($P = 0.041$) by the different diets, being predominantly higher in the COC group (2.33) and lower in the EOC and EOCCO groups (mean of 1.86). The levels of ash, total fat, and collagen were not affected ($P > 0.05$). The total MUFA content showed significant differences ($P < 0.001$) between treatments, being significantly higher in the CD group (57.36), intermediate in the COC, EOC, and EOCCO groups (mean of 55.79), and lower in the TPOC group (51.52). The total PUFA content was significantly higher ($P < 0.001$) in the TPOC group (12.21) and lower in the other groups. The same effect is observed in the PUFA/SFA ratio, with the highest content in the TPOC group and lower in the other groups. The SFA content did not show significant effects ($P > 0.05$). These results suggest that OC can be utilized in Bísaro pig feed without compromising the chemical characteristics and fatty acid profile of the meat.

In vitro metabolic footprint and interaction with intestinal cells of candidate probiotic and prebiotics for poultry

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Early microbiome modulation in birds can be managed already prior to hatch, by in ovo applications, but the bioactive compounds needs a careful validation prior to in vivo application. Metabolic footprint of candidate probiotics in intestinal cells in vitro was used to explain the molecular mechanism of their action in chicken intestinal cells. This study consisted of two parts: 1/ the selected candidate probiotic footprint was analyzed during culture with addition of selected prebiotics. 2/ the interactions of probiotic with intestinal cells were studied under co-culture conditions. To study the metabolic footprint, *Bifidobacterium lactis* NCC2818 was cultured at 37°C for 24 hours in a medium containing 2% [v/v] of two preselected prebiotic compounds. The supernatant samples (n=6 per each prebiotic) were subject to metabolites analysis by gas chromatography-mass spectrometry (GC-MS). Differential metabolite analysis showed changes in specific organic acids. The metabolic pathway enrichment analysis showed that metabolic pathways such as amino acid metabolism, energy metabolism, and two-component signal transduction systems were activated. Moreover, when interacting with intestinal cells, *B. lactis* was found to have a positive effect on the barrier function of intestinal cells. These results will be used to track the in vitro identified metabolic footprints in the in vivo study, in which the same probiotic was administered in ovo. The proposed in vitro methods can be practically used to explore and evaluate the potential candidate poultry probiotics, prior to in ovo application. The research was supported by National Science Centre Poland grant 2019/35/B/NZ9/03186(OVOBIOM).

Session 8

Poster 16

Effects of maternal antioxidant supplementation on the development of the offspring's ovaries

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Selecting good gilts is vital for farm productivity. A younger age at puberty correlates to better reproductive outcomes. This experiment aims to study if maternal treatment with antioxidants improves prepuberal ovary development of the offspring. Fifty Iberian sows were differentially supplemented from day 85 of gestation until weaning: control (C; 30 mg VE/kg feed); vitamin E (VE; 100 mg VE/kg); hydroxytyrosol (HT; 30 mg VE+1.5 mg HT/kg) and both antioxidants (VEHT; 100 mg/kg VE+1.5 mg/kg HT). Gilts were weighted at birth, during lactation and at weaning. At 110 days old, 10 gilts from each of the experimental groups were weighted and surgically castrated. The ovaries were measured and the number, size and density of follicles were studied histologically. ANOVAS were performed in R 4.3.2. Maternal treatment was used as a factor, whereas in repeated measures the gilt was a covariable in repeated measures. Gilt weights differed among treatments at birth (higher in VE and HT; p=0.078) and at day 20 of lactation (higher in HT and VEHT; p=0.05). Ovaries from VE gilts showed larger surfaces than the other groups (p=0.06), and thus had decreased follicle density (p<0.05). However, groups without VE supplementation had higher % of small follicles (p<0.05) that could imply lower ovary development. Thus, preliminary data suggest a long-term effect of offspring prepuberal ovarian functioning according to maternal antioxidant supplementation. The research was funded by the Spanish Agencia Estatal de Investigación.

Farm animal health and welfare at the heart of agroecological livestock system priorities: proposed research questions to contribute to this objective

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The aim of this communication is to summarize a collective, interdisciplinary vision of the advances in knowledge needed, at different scales, to contribute to the transformation of livestock systems with health and welfare at the core while considering the other components of sustainability. We emphasize the need to consider health in a broader perspective and to deepen the question of positive emotional experiences regarding welfare while presenting positive interactions and some conflicts between the two. Ex. when improving welfare by providing a more stimulating environment such as outdoor access the risk of infectious disease may increase. Jointly improving health and welfare of animals raises different questions along the production chain. At the animal level: long-term links between better welfare and physiological balance, the role of microbiota, the psycho-neuro-endocrine mechanisms linking positive mental state and health, and the trade-off between the physiological functions of production, reproduction and immunity should be explored. At the farm level, in addition to studying the relationships at the group level between welfare, health and production, we support the idea of co-constructing innovative systems with livestock farmers, as well as analyzing the cost, the acceptability and the impact of improved systems on their working conditions and well-being. At the production chain or territory levels questions include: studying the best strategies to improve animal health and welfare while preserving economic viability, labelling of products and consumers' willingness to pay, the consequences of heterogeneity in animal traits on the processing of animal products, the spatial distribution of livestock farming and the organization of the production and value chain. Citizens' knowledge of livestock farming should be improved and more constructive exchanges between breeders, livestock professionals and citizens developed. We emphasise a interdisciplinary and transdisciplinary approach, involving all stakeholders, including public decision-makers, in a participatory research process. Note: our numerous co-authors will be acknowledged during the presentation

Session 9

Theatre 2

Slow and local or fast and global – examining business opportunities to advance good welfare practices in organic and outdoor farming

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Although organic and low-input outdoor pig and poultry farming systems enable a high degree of expression of natural behavior by the animals, these farming systems still have challenges regarding their economic viability and practices that compromise animal welfare. Consequently, some of the variants of these systems go beyond the minimum standards of EU organic farming, and hence offer products of upgraded animal welfare. The objective of this study was to co-develop best practice business models that could be used to valorize welfare improvements in organic and low-input outdoor pig and poultry farming in Europe. The data for the study were collected by surveys and workshops organized in 9 European countries within the PPILOW project. The data were analyzed by using the business model canvas and the market opportunity navigator. In addition, costs, benefits and disadvantages of improvement options were examined. The value proposition is the central element of a business model and it provides guidance on how value is generated and how the selected approach is superior to alternative approaches. The developed business models focus on premium organic meat and eggs, which apply a set of welfare-improving measures that respond to ethical and animal welfare concerns related to pigs and poultry throughout their lifespan, enhance animal health, reduce animal antibiotic use and enable greater exploration of outdoor areas. Different strategies to enter the market were identified. The benefits and disadvantages of traditional and short supply chain were examined in the context of organic and low-input outdoor production. Novel revenue generation models such as subscription-based models and online-based sales will be considered. These approaches may involve features that increase the price to be asked from the market, but also make it more challenging to copy the business idea. The PPILOW project has received funding from the European Union's Horizon 2020 Research and Innovation Programme under grant agreement N°816172.

Behaviour and health of entire male pigs in a semi-natural habitat

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Entire male pigs in commercial housing systems are known for their increased agonistic and sexual behaviour. In wild boars, young males remain with the family group for about a year, then live in bachelor groups for approximately another year. The behaviour of domestic entire male pigs in family groups under semi-natural conditions has not been described to date. We therefore observed the behaviour of young pigs (Swiss Large White) in a 1.9 ha enclosure including pasture and forest. The social group consisted of three sows and their 41 piglets. Three months after birth, the group of piglets was reduced to 12. Pigs were observed once per week during four hours from an age of one month until slaughter at about 5.5 months. Our protocol included scan and continuous sampling through direct observations. We observed a higher frequency of head knocks between males or between males and females compared to between females. Fights were mostly observed between males, and rarely involved females. Mounting behaviour was performed nearly exclusively by males, with an observed drift in preference of the recipient: While in the beginning the recipients were similarly males and females, males mounted females significantly more than other males after three months of age. The prevalence of skin lesions was low and similar between males and females. In total 3 female and 3 male pigs were assessed as lame at least once. No penile injuries were observed at slaughter. These observations made under semi-natural conditions are in accordance with the described behaviour of entire males in housing systems.

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Theatre 4

Assessment of welfare of dromedary camels kept under pastoralism in Pakistan

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A protocol for measuring welfare in dromedary camels kept under pastoralism was developed, drawing from that one for dromedary camels kept in intensive systems. We aimed to apply the protocol and assess the welfare of dromedary camels in Pakistan. A total of 44 welfare indicators (animal-, resource, and management-based indicators) aligning with animal welfare principles ('Good Feeding,' 'Good Housing,' 'Good Health,' and 'Appropriate Behavior') were gathered into two assessment levels: 'Caretaker-Herd level' and 'Animal level'. Data were collected in 2023 in the Punjab province. A total of 54 herds/1186 camels, of which 510 (495 females and 15 males; average age: 5-6 years old) at Animal-level, were evaluated. The indicators were scored and aggregated to obtain Principle Aggregated Indexes (PAIs) and a Total Welfare Index (TWI). Using PAIs, 4 herds were classified as excellent, 42 satisfactory, 8 unsatisfactory. Good feeding and Good housing were the most problematic PAIs. TWI varied from 55.7 to 82.2, and using a traffic light classification, 12 herds were green, 22 orange, and 20 red. As expected, camels kept under pastoralism had a higher welfare than that reported in the literature for intensive systems. However, it was still possible to suggest guidelines for the improvement of those herds with criticalities. This is a first step to proposing welfare standards for camels.

Ensuring good animal welfare in extensive farming systems: lessons from dairy farmers in Canterbury, New Zealand

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Animal welfare is consistently identified as one of the most important, yet challenging issues for dairy farmers globally. Farmers in countries such as New Zealand, where systems are pasture-based and calving is seasonal, face a unique set of challenges associated with environmental stressors, access to adequate nutrition and disease management. To gain insights into current practices aimed at ensuring good animal welfare in extensive farming systems, in-person interviews were conducted with 5 dairy farm managers in Canterbury, New Zealand, in March 2023. The semi-structured interviews were based on 8 questions and lasted for 1-1.5 hours. The interviews were audio recorded and transcribed to facilitate thematic analysis. Five overarching themes emerged: “preventive herd health management”, “shelter”, “youngstock management”, “handling” and “feeding”. Under the first, farmers emphasized the importance of proactive measures like strategic grouping of cows and strict biosecurity to avoid introducing infectious diseases. They underscored the significance of early disease detection and the use of technological tools to facilitate this. Shelter was a recurring topic. While one farmer believed that cows could cope with all weather conditions, others stressed the importance of providing protection from adverse weather, particularly during calving. The discussion on youngstock management centered on collecting newborn calves, drenching routines and the lack of a male calf market. In terms of handling of cows, emphasis was placed on the necessity of well-managed walking tracks and the importance of employing patient staff to minimize stress during droving and milking. Lastly, issues around providing sufficient high-quality feed, particularly in the winter, were discussed. These findings provide valuable insights on the wide range of tasks performed to promote animal health and welfare on New Zealand dairy farms and add to our understanding of how producers using extensive farming systems can continue to adapt to increasingly strict welfare standards.

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Theatre 6

Organic pig production in mixed free-range systems: the ROAM-FREE project

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Background. Organic free-range systems combining pig production with pasture, cash crops, trees or other livestock may support self-supply of feed and biodiversity. However, such systems are challenging in terms of e.g. biosecurity, monitoring of animals, and land use. Objectives. Describe and analyze challenges and benefits in mixed free-range pig production across Europe. Methodology. The study included farms with different characteristics: Farms pigs + cattle and/or sheep (2 in Slovenia, 5 in Italy, 10 in Romania); Farms pigs on pastures with 10-30% tree cover (2 in Denmark); Farms pigs with cash crops and forest pasture (respectively 5 farms and 1, both in Norway) Farms were regularly visited, farmers interviewed, productivity recorded, welfare assessed and samples collected. Results. Outdoor pig rearing can provide improved animal welfare and increase job satisfaction. Farmers assessment viewed environmental benefits vague with pigs requiring careful management. Pathogenic, potentially zoonotic and resistant bacteria (e.g. E. coli, Pseudomonas) were shared between cohabitating sheep and pigs (SLO) but not amid sheep, cattle, and pigs (RO), welfare and immunity being influenced rather by raising pattern than cohabiting species. Pregnant and lactating sows were infected heavily with nodular worms while finishers had moderate levels of large roundworm (Ascaris), despite regular treatments (DK, RO). We expect that ROAM-FREE will reveal benefits and challenges in a variety of mixed systems and thus form basis for knowledge-based practical guidelines to improve existing free-range pig production.

On-farm animal welfare assessment in slaughter pigs across different production systems in four European countries

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In the mEATquality project (European Union grant no. 101000344), on-farm interviews and direct animal and system data collection was conducted to document aspects of animal welfare. Animal welfare assessments were performed on 80 herds across four countries: Denmark, Italy, Poland, and Spain (n=20 per country) and five production systems: Intensive, indoor, conventional (IIC, n=31); Intensive, outdoor run, conventional (IORC, n=4); Intensive, outdoor run, organic (IORO, n=11); Extensive, conventional (EC, n=24); Extensive, organic (EO, n=10). The herds were selected to reflect the variety of herds found in the respective countries with more intensive herds in Denmark and Poland, and more extensive herds in Italy and Spain. One major difference between the production systems was the amount and type of outdoor space with IIC herds by definition having no outdoor space, IORC and IORO herds mainly having access to concrete runs with a maximum outdoor space allowance of 1.33 m²/pig in Denmark and 193 m²/pig in Italy, and the EC and EO herds mainly having access to paddocks or pasture with a maximum outdoor space allowance of 53 m²/pig in Poland, 200 m²/pig in Denmark, 2,174 m²/pig in Italy and 84,000 m²/pig in Spain. Cooling systems also clearly varied with degree of extensification with intensive herds providing the pigs with sprinklers or showers, and extensive herds providing more natural cooling sources such as wallowing, river, ponds, pools, and shadow huts. Although the majority of herds provided no cooling facilities at all. Multiple other differences arising with extensification of the production will be presented at the conference, including differences between the production systems in resource and management indicators.

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Theatre 8

Navigating the complexity of boar taint detection: Advancing towards a castration-free future for piglets

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Most piglets are still surgically castrated in order to prevent boar taint, an undesirable odour, which can be noticed mainly during and after cooking. The two main compounds related to boar taint are androstenone and skatole, with existing analytical methods to detect them. While these methods are not feasible for boar taint detection on the slaughterline, the human nose (sensory method) is the most commonly used. One of its problems, is the accuracy and repeatability, since there is a biological variation in the chemical fingerprint that the human nose detects as boar taint. One objective of this study, as preparation for the PIGWEB TNA grant, was to examine the overlap between the objective and subjective measures and to create a combined model for boar taint detection. 4,000 boars were tested by the human nose on slaughter lines in commercial abattoirs; 2.1% of them were marked as tainted. All tainted backfat samples, as well as an additional 80 samples, were randomly chemically analysed and human nose tested by two additional trained panellists. The agreement between the two additional panellists was 24.3% only. 23.2% of all samples had androstenone or skatole above the selected sorting level (1500 µg/g and 250 µg/g, respectively). 5.0% of all samples had high levels of both. In total, only 3.4% of all samples had high androstenone, high skatole and were classified as tainted by the panellists. These results represent the importance of measuring both compounds as well as the untargeted chemical fingerprint. A novel model (based on advanced optics and AI) was created to take all these considerations into account.

Automated monitoring of pig and chicken welfare at the slaughterhouse – tracing back to farm, transport and slaughter

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The EU aWISH project focuses on automated monitoring of animal-based indicators at slaughterhouses for documenting the welfare of broilers and pigs during each phase of production (farm/transport/slaughter). The project is centered around six European pilots – a slaughterhouse with a local research partner, associated farms and technology providers – where technologies are developed and validated. Pig pilots are located in Austria, the Netherlands, Serbia and Spain, broiler pilots in France and Poland. Eighteen technologies measure various indicators for pigs (ear lesions, skin lesions, tail length, tail lesions, blood parameters, stress vocalisation, tear staining, stunning effectiveness, environmental parameters, body weight, lung lesions, liver lesions, food consumption, meat quality) and broilers (hock burn, footpad lesions, catching damage, back scratches, stress vocalisations, activity, stunning effectiveness, environmental parameters, on-farm welfare assessment). Technologies are mainly installed in the slaughterhouse, some on-farm or on-truck, and some require human observations. An overview will be given of the installation of technologies, their status of development and which animal welfare aspect they are measuring. Via a data platform and online user interface, feedback on relevant individual and aggregated indicator scores is provided to those responsible per production phase. Data will also be used to document animal welfare improvements and their socio-economic and environmental impacts. This project will make a significant contribution to the objective assessment and improvement of pig and broiler welfare throughout Europe.

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Theatre 10

Reliability of different behavioral tests as welfare indicator for dairy cows

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Human animal relationship (HAR) tests have the potential to reflect an important part of animal welfare on farm. To serve as welfare indicator, HAR-tests need to be reliable. Reliability of five different HAR-tests was therefore assessed by two trained observers who visited eleven commercial dairy farms in lower saxony. All farms were visited thrice, whereby visits were a week apart. At each visit, a novel object test (NOT: disinterested, interested, touched object, latency to contact), a voluntary human approach test (VHAT: latency to contact), a lie pass test (LP: disinterested, interested, stood up when passed) and an avoidance distance test (AD: flight reaction or touched by human, performed twice in different places) were conducted. Spearman correlation (rs) and intra class correlation (ICC) were used to assess the reliability of measurements between observers and between visits. Interobserver reliability of AD measurements were excellent (ICC and rS between 0.97 and 0.94). Reliability of NOT and LP categories varied from standing up (ICC and rS of 0.96) and touching the object (ICC 0.99, rS 0.98) to interest (0.92 and 0.90) in the NOT and (0.67 and 0.68) in the LP. ‘Disinterest’ was least reliable with rS of 0.45 in both tests and ICC of 0.65 in the NOT and 0.40 in the LP. Test-retest reliabilities were unacceptable (ICC and rS between 0 and 0.4). Low test-retest reliability can be explained by a small number of random animals chosen each visit. Exact influencing effects will be analyzed in the ongoing of the study.

Welfare protocols for rabbits

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In recent years, there has been a shift towards developing more standardized animal welfare assessment protocols, like the Welfare Quality® and the Animal Welfare Indicators (AWIN) projects. These projects have produced protocols aimed to be flexible and adaptable, allowing them to be used with different farm animal species. Nevertheless, due to a low economic impact or the limitations of its geographical distribution, the welfare assessment of rabbits in commercial production was left out of both projects, even though this species are primarily reared in intensive systems. In 2011, there's a first attempt at creating a welfare protocol for on-farm commercially housed rabbits, through observation and scoring methodologies. This first approach was mainly based on the Welfare Quality project and available literature, as well as expert knowledge. Several authors have adapted this work ever since, which is now mostly used to evaluate farms in Spain and Portugal. In 2022, the EBENE protocol for does and growing rabbits is introduced in France, following the same principles but with more practical presentation and approach. At the same time in Italy, a protocol based on Classyfarm is developed. Most of the recent versions of welfare assessment protocols for rabbits have a lower number of indicators in common and a lack or severe decrease in behavioural assessments, which poses an obvious problem. There is still a need for more research in indicator validation, feasibility and repeatability. In this work, we analyze and compare each protocol proposal recently designed for rabbit production. This work was supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

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Theatre 12

Behaviour of laying hens of two genotypes in a cage-free system enriched with hay bales

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Environmental enrichment can play a positive role in cage-free systems by allowing hens to diversify their behavioural repertoire and reducing aggression among animals. Our study evaluated the effect of the enrichment with hay bales in an aviary system on the behaviour of 1,800 laying hens (Hyline Brown and Lhomann White) from 50 to 53 weeks of age. Behaviours, expressed as the percentage of hens observed on the floor, were scored through a 10-sec scan every 30 min per hour (05:30-20:30) and observations were grouped per day-time interval, i.e. early, midday, afternoon, and evening. Data were submitted to ANOVA with genotype, enrichment, and time interval (not discussed here) as main effects, and pen as a random effect. As for the enrichment, the presence of hay bales increased the number of hens on the floor and the rate of those interacting with the enrichment at the expenses of hens performing dustbathing, sitting, or walking. As for the genotype, a higher number of brown hens was recorded on the floor, with a higher rate of them sitting-inactive and preening compared to white hens which performed more dustbathing, interactions with hay bales, ground-pecking, and piling ($P < 0.01$). In conclusion, hay bales attracted hens and reduced the expression of some comfort behaviours; white hens were more active and showed more risky behaviours such as piling compared to brown hens. Acknowledgements: This study was carried out within Agritech National Research Center and was funded by the European Union – NextGenerationEU (PNRR – Missione 4 Componente 2, Investimento 1.4 – D.D. 1032 17/06/2022, CN00000022), and by Unimpresa, University of Padova (call 2019).

Improvement of the welfare of Iberian cull sows fattened through the immunocastration

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To enhance the sustainability of Iberian traditional pig production, it is suggested to fatten Iberian cull sows using the natural resources of the dehesa ecosystem. However, during the fattening period, hierarchical aggressions between entire sows may occur due to oestrus, as well as unwanted pregnancies by wild boars in free-range systems. To mitigate these issues, this study aimed to evaluate the performance and welfare of entire and immunocastrated Iberian cull sows during the fattening period. The study was conducted on 36 multiparous Iberian sows at the CICYTEX experimental farm. One group of 18 sows remained entire, while the other group of 18 sows was immunocastrated using Vacinsinzel® before the fattening phase. After the immunocastration protocol, both groups were fattened in the dehesa using natural resources. The animal welfare of both groups was assessed every five weeks (4 assessments) using the Welfare Quality® protocol. Monthly body weight (BW) measurements were taken, and blood samples were collected for analysis of progesterone and estradiol (ELISA). Statistical analyses were conducted using SAS®. Regarding animal welfare, differences were only observed between the two experimental groups at the end of the fattening period according to the Welfare Quality® protocol. Expression of positive social behaviours was found to be more frequent in immunocastrated Iberian cull sows. Analysis of progesterone and estradiol confirmed the effectiveness of immunocastration. There were no significant differences in BW and average daily gain between the two experimental groups. In conclusion, immunocastration is an optimal management tool to improve the welfare of Iberian cull sows.

Session 9

Poster 14

Impact of outdoor farming on pig welfare: insights from a study in Italy

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The current scientific position on animal welfare promotes farming systems that allow the free manifestation of the species ethogram. For pigs, access to the open air and to grazing areas represents an added value in terms of welfare as a place in which they can express their rooting behavior. This study focuses on five Italian organic farms where pigs are raised outdoors, and grazing is a part of the daily ration. The aim was to investigate how farms organized grazing and free-ranging activities in relation to animal welfare assessed using the app PIG LOW. Furthermore, we have identified the risk factors for parasitic infestation connected to each situation and the related parasitic condition of the animals. The objective was to obtain information on management practices that promote animal welfare and to understand whether and how these influence parasitic infections in pigs. The study was conducted as part of the European ROAM-FREE project in spring 2023. The PIG LOW app was used to evaluate various aspects of animal behavior and environmental conditions during visits to five Italian organic farms where pigs were raised outdoors. This method allowed them to objectively evaluate animal welfare and understand the management practices in place. The results indicate that outdoor farming systems can enhance the welfare of pigs by enabling them to engage in their natural behaviors. Despite the higher risk of parasitic infestation in outdoor environments, the study suggests that appropriate management practices can effectively mitigate these risks. Another fundamental aspect was the pigs' confidence with humans. In most farms, the animals did not exhibit fear towards humans: this shows that the animals feel relaxed and safe, without any sense of danger, which is an excellent outcome from a welfare perspective.

Could a low input/low output forage-based system be the solution for future organic dairy production? An exploration of the concept of a LILO production system

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Dairy production has been strongly affected by increased prices for inputs, such as energy, fertilizers and high-quality concentrate feed. Meanwhile, the demand of organic dairy milk has been on the decline since 2016, which has led to a reduced financial return for organic producers. Organic dairy production is normally more costly than conventional, as the inputs usually are more expensive due to the need of them to be certified as organic. As the feeding costs account for the largest part of the inputs in dairy production, it is important to explore alternatives. By implementing a forage based, low-input/low-output (LILO) strategy in organic dairy production, the costs of input would be lowered, mainly by decreasing the use of concentrate feed and replace it with high-quality roughage combined with home-grown grains. This would not only preserve organic production, but also potentially benefit animal health and welfare as well as several crucial ecosystem services and leave more room to grow human-edible feed for humans. The first step is to untangle what the concept of a LILO system is in a Swedish perspective, in terms of the composition of a herd's feed ration. We will explore this by using existing information on forage consumption, type of forage and grazing at farm level. This is a part of a larger research project on how LILO systems affect animal health and welfare, farm economy and the climate footprint.

Session 9

Poster 16

First evaluation of the CLASSIFYFARM welfare assessment protocol applied in extensive farming systems in Central Italy

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According to European regulations and the Animal Health and Veterinary Drugs Directorate of the Italian Ministry of Health, Classifyfarm serves as a welfare assessment protocol for categorizing livestock farming based on risk levels. On-farm assessments encompass biosafety, farm management, barn structure, animal-based measures (ABMs), as well as hazard and alarming systems. To assess the strengths and weaknesses of extensive rearing in Central Italy, we conducted evaluations on 20 farms situated in the agroecological systems of the Lazio region. These farms primarily raise autochthonous cattle, horses, sheep, and goat breeds. The results offer crucial insights into farm structure and management. Notably, the biosafety section poses challenges for small-scale ones due to their limited herd size and building capacity. Consequently, we emphasize the need for a more tailored questionnaire within the Classifyfarm system, accommodating the unique characteristics of extensive farming. This adaptation is essential for drawing appropriate conclusions about overall categorization. This necessity becomes more pronounced given the association of Classifyfarm usage with public subsidies (Ecoschemes, the national label SQBNA, regional Rural Development Programmes), and the potential future mandate for its implementation. Such a requirement may pose a significant challenge for small-scale and extensive agricultural activities, potentially resulting in the abandonment of these practices. This, in turn, could lead to the loss of substantial local economies and associated ecosystem services. Funding: Activity carried out with the contribution of the Rural Development Program (PSR) Lazio 2014-2022, Operation Type 10.2.1 'Conservation of plant and animal genetic resources in agriculture,' CUP: F85E22000480009.

Season effects on somatic cell count as an indicator for mastitis in Romanian Spotted cows

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Udder health is of high importance for both animal welfare and for economic considerations in regard to milk quality and quantity. The aim of this study was to establish the season pattern during a three-year period of somatic cell score (SCS) in the first 3 lactations of Romanian Spotted cows. A total number of 376 animals with 4429 test-day milk records were used. The values of somatic cell count (SCC) were recorded every 28 days and then transformed to SCS. To determine the season pattern on SCS, a linear mixed effects model was built incorporating pedigree data, using the “lme4qt1” R package. Parity (lactation rank), days in milk, year and season of test day were included as fixed effects, while the animal was used as random effect. Cows in the second (L2) and third lactation (L3) had a significantly higher SCC compared to those in first lactation (L1), resulting in an increase of 0.49 units in SCS between L1 and L2 ($p < 0.01$) and 0.75 units between L2 and L3 ($p < 0.01$), suggesting that higher lactation ranks are associated with higher risks of mastitis. A significant effect of season was observed on SCS and mastitis frequency ($p < 0.05$). Thus, spring was associated with an increased ($p < 0.01$) SCS by 0.25 units compared to summer and by 0.23 units compared to autumn. Also, summer and autumn had higher frequency of healthy cows compared to spring (63.2% and 60.4% vs. 56.2%). There was no significant effect of winter on either SCS and mastitis frequency. These results show an effect of season on SCS. This research was funded by the project ADER 7.1.9/2023.

Session 9

Poster 18

Environmental enrichment in rabbit generations: Impact on behavior and performance

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Pre and post-natal life experiences of offsprings may impact adulthood and subsequent generations phenotype. Here, we assessed the impact of environmental enrichment during the pre and postnatal phases of rabbits' behavior and performance. In a 2×2 factorial experiment, 16 New Zealand rabbit does were randomly allocated into four treatment groups. During the prenatal phase (0 to 27 days of age), and lactation phase the does were housed in either enriched or conventional cages resulting in four experimental treatments: enriched-enriched (E-E), enriched-conventional (E-C), conventional-enriched (C-E), and conventional-conventional (C-C) housing. Conventional cages measured 80 x 60 x 40 cm, while enriched cages were eight times larger that included hiding spots, two-floor levels, vertical stretching space, and box with wood shavings. After weaning, all kits were moved to conventional cages. For performance we evaluated litter size, number of weaned kits, birth weight, and weaning weight. Two female kits per litter were randomly selected and tested on open field, novel object, and social interaction tests at 90 days of age. The results indicate that only the enrichment during the postnatal phase influenced the outcomes. C-C had on average 8.2±1.2 kits per litter, while E-E had fewer kits (4.2±1.2; $P = 0.03$). C-E and E-C did not differ from C-C. Weaning weight and birth weight showed no differences among the treatments. There were differences in the open field and social interaction tests for C-C compared to C-E but not compared to others. C-C explored less quadrants (15.4±3.01) and had fewer fights (0.0±0.83) compared to C-E (20.9±2.96; $P = 0.07$) and (3.5±0.82; $P < 0.01$), respectively. Therefore, results suggest environment enrichment in postnatal phase impact on behavior and performance of rabbits.

Animal welfare: perceptions and practice evolution in the broiler production in France – A pilot study

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Over the last decades, animal welfare has become a major societal issue, especially regarding farm animals' breeding conditions. Production systems, in particular those involving highly impoverished environments, must evolve, and breeders are facing new challenges. In France the standard poultry production and the inherent restricted breeding conditions is under scrutiny. As part of a larger research project about the improvement of animal welfare in standard poultry production, the aim of this study was 1- to determine how breeders perceive the interrogations around animal welfare and the improvement propositions, and 2- to identify the improvement strategies they implemented on farm. Semi-directive interviews were conducted with 26 French breeders to characterise the farms, determine their knowledge and interests in terms of animal welfare, identify their perception of the solutions proposed to improve broiler welfare and identify their welfare improvement strategies. Most breeders (76%) had a negative perception of the institutional discourse around animal welfare, mainly because of the lack of economic valorisation of animal welfare friendly practices (38% of the breeders). In contrast, the practical actions aiming at improving animal welfare on-farm were mainly positively perceived, from the participation to professional training to the introduction of enrichment items (e.g. straw bales, pecking objects) (46%: "important", 21%: "improved image"). All the breeders involved in our study included at least 1 enrichment object in their facilities in the last year, and 61.5% included 4 to 5 enrichment items, including an access to daylight. This preliminary study offers a positive insight on the breeders' willingness to address and take over the animal welfare subject, when properly supported.

Session 10

Theatre 1

Livestock are more than food: welcome and introduction

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This joint Animal Task Force – EAAP Livestock Farming Systems symposium explores the non-food contribution of livestock. In most farming systems, livestock's main function is food production, but they provide many other products and services such as fibre, hides and skins, energy, manure for fertilisation, feed, insulation of houses, functional components beyond basic nutrition, compounds used in medicine (e.g. insulin, heparin and hyaluronic acid), biodiversity and landscape management (e.g. fire prevention), traction, and contributions to culture, mental wellbeing as well as being an integral part of vibrant rural communities across most of Europe and beyond. In the context of climate change, measurement of the use of animal resources in all its components is a key question, and it is important to take all the products and services provided by livestock into account when assessing their contribution versus their environmental impact. The multiple products provided by livestock limit the value of comparisons with other foodstuffs using single factor assessments as often carried out in life cycle analysis exercises. All these non-food uses, in addition to food, contribute to the closing of the biological cycle, and are key points to reduce waste and valorise the contribution of animal production to the circular bio-economy. Some of them, like manure and drug production are alternatives to the use of fossil resources and contribute significantly to improving the climate balance of animal production. Animal by-products also contribute significantly to land use efficiency where alternative plant-based products would utilise land which could have otherwise been used to produce food directly (e.g. processed animal-protein versus soya). The theme covers all the species and involves all the actors including production, industry, and the co-product valorization sector. The topic aligns closely with EU policies such as the Green Deal and Farm to Fork strategy, the Bio-economy Strategy, Animal By-products legislation, food waste reduction targets, and the Sustainable Food Systems framework.

Role of livestock in circular bioeconomy systems

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Adopting circularity is key to sustainable food production systems. Livestock are increasingly scrutinized for their greenhouse gas emissions and broad environmental footprint, with their role within the circular bioeconomy often overlooked. To address this, the FAO Livestock and Environmental Assessment and Performance Partnership program convened a panel of 36 experts from 24 countries to develop guidelines to integrate circular bioeconomy principles into the environmental assessment of livestock supply chains. Life cycle assessment and food systems modeling can assess the circularity of livestock systems, but they do not consider all possible indicators. Livestock promote circularity by utilizing plant-based by-products from food and plant oil processing as feed. If not feed, many of these by-products would likely be combusted as fuel, biodigested or landfilled. Besides milk, meat and fibre, livestock produce an array of co-products used in the pharmaceutical, cosmetic, textile and chemical industries. Rendered co-products from meat processing are valuable sources of energy, protein, and minerals. Bio-energy technologies can capture energy from manure and other residues, and recycle organic matter and nutrients back to soils. Circular bioeconomy policies and regulations are key to ensuring that circularity practices offer an overall benefit to food supply chains. These practices need to be assessed regionally so that livestock production practices promote circularity in a manner that minimizes the environmental footprint while optimizing the delivery of ecosystem services from agricultural systems. Besides, assessing these practices will generate the data needed to inform the development of livestock food systems through a circularity lens.

Session 10

Theatre 3

Beyond edibles: unveiling the full socio-economic value of animal production through non-food products in circular economy designs

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The increasing awareness of the environmental footprint of livestock farming, alongside concerns regarding animal welfare and the health implications of excessive meat consumption, has galvanized a reevaluation of dietary norms globally. As the urgency to mitigate these impacts grows, the quest for sustainable dietary alternatives to meat becomes paramount. This research proposes to explore the comprehensive effects of transitioning towards diets with reduced meat content, evaluating not only the environmental but also the economic, social, and cultural dimensions of sustainability. A critical but often overlooked aspect of reducing meat consumption is the circular economy engendered by livestock farming by-products. For instance, animal manure plays a vital role in agriculture, serving as an organic fertilizer, while the leather industry, highly reliant on animal hides, underscores the economic significance of meat production by-products. Cultural factors also play a pivotal role in dietary choices. The challenge lies in balancing respect for cultural heritage with the imperative for sustainable consumption patterns. As alternatives to meat gain traction, understanding their potential impacts on cultural practices becomes crucial. Moreover, the sustainability impacts of meat alternatives across their life cycles remain under-researched. Questions persist regarding the environmental, social, and economic ramifications of scaling alternative protein sources. For instance, the cultivation of certain crops for alternative proteins could entail significant land use, water consumption, and possibly, the displacement of traditional farming practices. This research aims to offer a comprehensive analysis of the costs and benefits associated with reducing meat consumption, drawing on the latest research to map out the multifaceted dimensions of sustainability involved. By examining the economic circularity stemming from livestock by-products, alongside the social and cultural implications of dietary shifts, the goal is to present a holistic view of what a sustainable dietary future might entail.

Genetic determinism of quality of lamb leather in Lacaune dairy sheep breed

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Enhancing the economic valorization of by-products from dairy farming, such as skins and wool, serves as an important element in ensuring the economic sustainability of sheep farming. However, achieving this improved valorization necessitates a comprehensive understanding of the factors influencing the quality of these by-products. Lacaune lamb skins have long been esteemed for their exceptional quality. Over the past few decades in France, however, the quality of this leather has declined, primarily attributed to the increasing prevalence of two skin defects: pinhole and straw-like bushiness. To explore the genetic determinism of these defects and their potential correlation with wool traits, approximately 1400 Lacaune lambs from 72 rams underwent phenotyping for the two skin defects and four wool traits. The pinhole defect exhibited a high heritability (0.59 ± 0.07) and demonstrated a strong genetic correlation with the predicted core bulk of the wool ($rg = 0.54 \pm 0.08$). The straw-like bushiness defect displayed a lower heritability (0.25 ± 0.06) and was found to be correlated with the mean fiber diameter ($rg = 0.41 \pm 0.15$). In an initial study, the two skin defects exhibited either null or only weak genetic correlations, most often favorable, with milk production traits (milk yield, fat and protein contents). A genome wide association study revealed for the pinhole defect one major signal on chromosome 3 close to a cluster of keratin genes.

A systemic description of the dairy value chain's contribution to inclusive sustainable development in highland

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The present research, conducted in the framework of the HIGHLANDS.3 project (H2020, MSCA-RISE) aims to investigate on the benefits of the dairy value chain beyond food provision, and their contribution to inclusive sustainable development in highland. A transdisciplinary team interviewed 15 cases related to the dairy value chain across four highland regions in Europe: Alps, Carpathians, Pyrenees, and Massif Central. Actors interviewed were farmers, cooperatives, retailers and associations. A system thinking approach was adopted to process data. For each case were identified the factors contributing to inclusive sustainable development in highland, then clustered within similar topics. Then, a comparative analysis identified commonalities and differences between the cases. A causal loop diagram (CLD) was constructed for each region and another CLD integrating the regional data. The CLDs were then interrogated to identify subsystems and potential points of leverage, to enhance the sustainability of the system and to identify differences and similarities across the regions. The results indicate that dairy value chain in mountain areas provides multiple benefits beyond food production. They include enhanced ecosystem services, strengthened local communities, added value products, protected local resources and strengthened farm families. Activities like cooperation, diversification of activities and local commercialization contribute to strengthen the local community. Differences between the cases included traditional practices (transhumance), re-use of by-products and action to reduce the environmental impact.

What is the performance of a low-input sheep flock integrated into an agroecological field crop system?

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The association of animals and crops is a way of strengthening the sustainability of farming systems. For two years, we tested the association of 180 ewes with field crops grown without synthetic pesticides, on an experimental farm. The most fundamental point was to graze the intercrops planted after crops had been harvested. To achieve this, lambing was planned in September, and the great majority of lambs were fattened exclusively on these resources from November until their sale in April. The flock was kept in complete free-range conditions. Productivity results were high, with an average 1.45 ewe productivity and lambs carcass weight rather high (18.4kg). The use of concentrates (both for ewes and lambs) was limited to 3.4 kg per kg of carcass produced, compared with 9.6 kg for conventional sheep-crop farming systems. This technical performance translates into high economic and environmental performance: €126 gross margin per ewe (vs. €95 in reference), 24.3 EqCO₂/kg carc. (vs 31.6 in ref), and 65 MJ/kg carc. (vs 92). However, this performance could be improved as it is penalized by two points: the relatively high level of nitrogen fertilization, in view of crop yields, and the high proportion of lambs reared on artificial milk and then fattened with concentrates (24% of lambs), linked to the very high prolificacy. We consider these results to be promising. Not only does this system deliver excellent economic and environmental performance, it also meets the demands of the industry, with sales at the end of winter. However, we still have to overcome the cultural and organizational obstacles associated with setting up sheep flocks on specialized crop farms.

Session 10

Theatre 7

Livestock manure for fertiliser and biofuels

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Livestock manure is a very important resource of plant nutrients and contains large amount of organic material that can be converted to energy and fertilizer (digestate). At the same Livestock manure is a major source of the greenhouse gases (CH₄ and N₂O) and a source of nitrogen and phosphor pollution. The emissions can be mitigated by production of biogas through anaerobic digestion (AD) of manure, mostly together with other biowastes, which can substitute fossil energy and thereby reduce CO₂ emissions and postdigestion GHG emissions (Møller et al. 2022). The nutrient value in digestate can be improved and replace significant amounts of mineral fertilizers and improve soil fertility. In countries like Denmark almost all biogas plants use co-digestion and the biomass used for AD is highly variable between biogas plants. Almost all AD plants use around 75% of the biomass as manure. The feeding of animals and the stable system plays a large role for the quality of the manure for AD and the environmental impacts. Furthermore, additives like lipids used to reduce enteric fermentation in dairy cows plays a role for the energy yield and environmental impacts (Møller et al. 2014). The digestate products and its value as fertilizer depends on the biomass used and the biogas technology. In general, the fertilizer value can be improved by AD but the ammonia emission potential of digestate applied in the field is higher than that from cattle slurry and pig slurry because of the higher pH of the digestate. However, there are several technologies available to reduce losses of ammonia. The current knowledge on energy potential, climate impact and nutrient value of digestate from different types of animals, feeding of the animals and biogas technologies will be presented. References: Møller, H.B. Verónica Moset, Maike Brask, Martin Riis Weisbjerg, Peter Lund, 2014. Feces composition and manure derived methane yield from dairy cows: Influence of diet with focus on fat supplement and roughage type. *Atmospheric Environment*, 2014. 94: p. 36-43. Møller H.B., Sørensen P, Olesen JE, Petersen SO, Nyord T. & Sommer SG, 2022. Agricultural biogas production – climate and 225 environmental impacts. *Sustainability* 14, 1849.

Cow's Milk – Going beyond basic nutrition to harness functional components

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Milk protein consists of ~80% casein and ~20% whey proteins. These whey proteins include alpha-lactalbumin, beta-lactoglobulin, lactoferrin, glycomacropeptide, serum albumin, lactoperoxidase, immunoglobulins and other components that possess a variety of different health promoting functionalities, i.e., go beyond basic nutrition. The functionality of milk can be further optimised through processing to enrich some of the aforementioned functional components in whey, through enzymatic hydrolysis of milk to reveal new functionalities (such as cryptic peptides) or fermentation (generating yoghurt, kefir and other potentially health promoting products). Associated benefits arising from the consumption of such functional ingredients can include, for example, the control of weight, appetite, inflammation, hypertension, stress as well as prebiotic effects and benefits arising from modulation of the gut microbiome.

Session 10

Theatre 9

Evolving standards: ethical perspectives on animal welfare in agroecology

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Animal welfare (AW) is a crucial aspect of the EU's Farm to Fork strategy, which seeks to expedite the shift to sustainable food systems through integrated food policies. Agroecological farming encompasses extensive, circular, and mixed farming models that safeguard biodiversity, recycle nutrient, regenerate, and preserve local resources and knowledge, and promote a holistic interaction between nature, animals, and humans. However, there are still many challenges to overcome. For instance, horizontal legislation lacks a clear definition of "factory farming" and there is a shortage of EU standards to ensure the AW of farmed animals. The current situation is impeding the transition phase and creating uncertainty in a livestock system that is frequently in crisis, and characterised by ageing, abandonment, and low economic marginality. Attempts have been made to bridge this gap, but often through the implementation of top-down measures that aim to standardise and unify biodiversity, resulting in detrimental spillovers on the safeguarding of heritage breeds and small-scale farms particularly in marginal areas. This approach of imposing "one size fits all" solutions should be avoided, and a more nuanced and context-based approach is needed. Coherent, and effective examples of policies tailored to the "farmers' first" concept have to be followed more closely, e.g. i) a new evaluation system where "animal-based" parameters would become the cornerstone of an on-farm AW assessment in different farming contexts, and in all regions; ii) a participatory approach to livestock selection with a direct animal assessment carried out by farmers for qualitative traits could shift breeding goals towards functional rather than production traits; iii) fostering technical solutions able to represent agroecology as productive and profitable food production system even without requiring massive subsidies: a transition from sustainable intensification to sustainable extensification of agriculture is needed, through a process of ecological increase; iv) a large scale deployment of the "One Welfare" approach as an horizontal framework to promote AW in connection with human welfare, social justice, and environmental conservation.

Session 10

Theatre 10

Panel discussion

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Panel discussion with speakers and audience

Session 11

Theatre 1

Growth performance of black soldier fly (*Hermetia illucens*) reared on substrate mixtures from regional agricultural and food production side-streams

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Larvae of the black soldier fly (BSFL) can convert feed and food residues into high quality protein and lipids. The aim of this study was to develop biomass residues mixtures (BRM) from regionally available plant and animal residues. The BRM, named after the main protein source (high fish (HIFI), low fish (LOFI), *Silphium perfoliatum* (SIPE), pork slaughter (SLRE) and canned fish (CAFI) residues) were nutritionally characterized and contained 14-21% CP, 8-15% CL and 22% NDF. The BRM were adjusted to 30 % DM and growth performance was compared with a positive control (chicken feed, CF) and a standard fly diet (Gainesville diet, GD). All substrates were fed (5 kg FM/box (40*30*17cm), n=6, N=42) to 5-day-old BSFL (5dof) until first prepupa were present. Then nutrient contents of BSFL and frass were determined and data were analysed statistically (Proc mixed, Tukey-Kramer, SAS). The fattening period was longest for SLRE, CAFI, SIPE (18 d), intermediate for HIFI, LOFI, CF, and shortest for GD (14 d). The numbers of larvae harvested for CF, GD, HIFI, LOFI, SIPE were similar to numbers of 5dof. Individual body mass was lowest for GD (113 mg), intermediate for BRM (HIFI 178 mg), and highest for CF. Larvae mass/box was in ascending order SLRE, CAFI, GD, SIPE, LOFI, HIFI, CF (951 g/box). Overall, CF performed best, followed by HIFI and LOFI. Surprisingly, growth on SIPE was better than on CAFI and SLRE, despite of lower CP. Concluding, nutrient digestibility and physical properties of the BRM could explain the differences, but this requires additional research. Funding: Federal Ministry of Education and Research, FK 031B0915L6.

Fortification of circular substrate for black soldier fly with zinc and/or selenium: effect on growth performances

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Zinc (Zn) and Selenium (Se) are essential trace elements for livestock. To ensure an adequate intake is achieved, several supplementation sources have been studied. Emerging and innovative sources, such as Zn and Se-enriched insects, could be useful for their potential use in animal nutrition. The purpose of this work was to study the effects of including Zn and/or Se in the rearing substrate of black soldier fly larvae (BSFL), where a lack of information regarding growing performance effect was identified. In this study, BSFL were reared on five different substrates: a control substrate (CTR diet) of plant-ingredients (Gainesville diet); Okara and Potato Waste based substrate (OPW); OPW-Zn fortified with 150 mg/kg of Zn; OPW fortified with 0.3 mg/kg of Se; and OPW-Zn+Se fortified with 150 mg/kg of Zn + 0.3 mg/kg of Se. All experiments were carried out under dark condition, at 26 °C with 60% relative humidity and the growth performance of the resulting larvae and prepupae were observed. The mean larval weight was not influenced by treatment ($P>0.05$). In contrast, the CTR group showed significantly higher larval biomass yield at the end of the trial ($P<0.05$). The number of resulting larvae and their survival rate were not influenced by the treatment ($P>0.05$). These results suggest the possibility of use these substrates. However, more studies are necessary to determine the effects on reproductive performance of BSF adults as well as their proximate composition. Funded by Agritech National Research Center, spoke 8

Optimal balance between protein and carbohydrate in industrial diet for black soldier fly larvae (*Hermetia illucens*)

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Black soldier fly larvae (BSFL), *Hermetia illucens*, can efficiently convert organic substrates into insect biomass, which is suitable for livestock feed. The performance of BSFL depends on the macronutrient composition of their diet, and especially on protein (P) and carbohydrate (C) content. This study uses a geometric approach to determine the effect of different ratios between P and C concentrations in semi-industrial diets formulated with cereals and tubers by-products typical from North-East of France. Seventeen oligidic diets varying in their P and C concentrations were formulated, with C+P concentrations ranging from 25% to 65% and C:P ratios from 1:1 to 4:1. These diets were used to feed the BSFL for seven days. The impact on the feed conversion ratio (FCR), protein and lipid content of 14-day-old larvae (14 DOL) was assessed. Initial feed provided and frass were also analyzed to account for estimated digestibility of P and C. Our results indicate that BSFL performance is more affected by C+P concentrations than C:P ratios. The best-performing diets for minimizing FCR were those with a C+P concentration superior to 45%, while the 14 DOL protein plus fat content was maximized with diets containing approximately 50% C+P and a C:P ratio between 2:1 and 4:1. These findings complement existing literature by studying semi-industrial diets with a narrower range of P and C, and accounting for macronutrient digestibility. They confirm the significance of the diets P and C content in optimizing BSFL performance at an industrial scale. Future research should explore optimal ratios different types of carbohydrates and amino acids to further optimize BSFL performance.

Evaluating amino acid requirements of neonatal black soldier fly larvae

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Amino acid (AA) requirements of black soldier fly larvae (BSFL) during the early nursery phase remain poorly understood. We conducted a feeding trial including a semi-artificial low-protein nursery diet (NC) that was complemented with 20 free AA to reach 18% protein content (PC) and to simulate the AA profile of a commercial laying hen feed (RC). In 12 additional treatments, relative to PC levels, either Arg, Ile, Lys, Met, Thr, or Trp contents were reduced by 31-46% via leaving out respective free AA or individually supplemented to 131-146% (-AA vs. +AA treatments). Each treatment was triplicated with 10,000 freshly hatched (naïve) BSFL and restrictively provisioned with 10 mg dietary dry matter per larva. Growth was most delayed with NC and -Lys. Conversely, -Met and -Trp responses were similar to PC while growth curves for -Arg, -Ile and -Thr were intermediate. Adding Met, Thr, Trp, and Ile did not improve performance over PC, but +Lys and +Arg both steeped growth, resulting in fastest readiness for fattening. All single -AA treatments needed 1-5 days more than respective +AA treatments to reach critical larval live weights (~10 mg). For the actual basal diet, we conclude that neonate BSFL growth is first limited by Lys and co-limited by Arg, Ile and Thr, whereas Met and Trp seem indifferent.

Session 11

Theatre 5

Amino acid requirements of the black soldier fly larvae (*Hermetia illucens*)

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The nutritional requirement of black soldier fly larvae (BSFL; *Hermetia illucens*) is important for optimizing production of BSFL and assess the applicability of different feedstuffs based on chemical content. However, little is known about the amino acid (AA) requirements. The current study explored if the AA profile of chicken feed is the optimal for BSFL. The optimal AA concentrations were tested for ten AA that are essential for mammals: Met, Lys, Ile, Leu, Phe, Thr, Trp, Val, Arg, and His. A diet containing chicken feed, carbohydrate mix, cellulose, crystalline AA mix, mineral mix, and agar served as basal diet with 15.2% crude protein. The AA profile from the crystalline AA mix resembled that of chicken feed, except for the AA to be evaluated. Experiments took place in a climate chamber with a temperature of 27±1°C and relative humidity of 55±10%, in 370 mL plastic containers, containing 100 g experimental diet and 123 larvae. Larvae were 6 days old at the experiment start, and the experiment lasted for eight days. Each treatment had seven replicates. A dose-response experiment was carried out on each of the ten AA, investigating growth and survival at seven different concentrations of the AA. Addition of Met, Arg, and His did not improve larval growth, indicating the lowest tested concentration of 1.75g Met, 5.12g Arg, and 2.00g His/kg DM were sufficient for optimal growth. Only for Thr, larvae exhibited greater weight gain when exposed to a concentration higher than those currently found in chicken feed (6.49 g/kg DM). Val supported optimal larval growth at concentrations equal to chicken feed (7.62 g/kg DM). For Lys, Ile, Leu, Phe, and Trp, the concentrations resulting in highest larval weight was lower than in chicken feed (6.99 g/kg DM, 5.72 g/kg DM, 8.01 g/kg DM, 5.04 g/kg DM, and 1.47 g/kg DM, respectively).

Growth performance of black soldier fly larvae on high fibrous residual streams pretreated by composting and fungi

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High volumes of high-fiber residual streams are often used in biomass energy plants. Alternatively, these could be used in substrates to grow black soldier fly larvae (*Hermetia illucens*; BSFL). Therefore, this study aimed to assess the growth performance and safety of BSFL reared on reed, grass, Japanese knotweed, and nature straw after composting and inoculation with fungi. Pilot experiments were performed to select the most promising combinations for a large-scale study. In pilot 1 growth performance was determined in 41 combinations of residual streams with 50 BSFL per treatment (25 g of substrate; n=1). Composted nature straw whether or not with fungi, and composted reed, grass, and Japanese knotweed were the most promising treatments and selected for pilot 2 with 200 BSFL per treatment (100 g of substrate; n=3). Survival of the larvae was close to or over 90%. The yield was the highest with composted nature straw as a substrate as it resulted in an average fresh larval weight gain of 26 mg in a six-day larval period. The growth performance of the tested residual streams was low compared to chicken feed as reference substrate (larval weight gain of 111 mg), indicating a low nutritional value. In follow-up experiments, the additional value of the side streams will be tested as a structure ingredient in substrates consisting of chicken feed to study possible stimulating effects on the growth performance of BSFL.

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Theatre 7

Fat chemical characteristics of the Black soldier fly larvae fed different diets

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Black soldier fly larvae (BSFL) can transform organic wastes into an enriched nutritional biomass useful in animal feeding. So, BSFL may be useful to increase sustainability of animal derived food. However, little information is available about the effect of diet on the fat nutritional value of BSFL. The aim of this work was to study the effect of five diets (based on meat or fruit or vegetable substrates or a mix of them and control) on fatty acid (FA) profile of triglycerides, phospholipids, and sterols. The dietary effect was estimated on two BSFL populations obtained in two different facilities located in Greece (GRE) and Italy (ITA)UNIFI. The diet affected all the lipid fractions examined. Independently of diet, the larval fat was characterized mainly of lauric acid and other saturated FA. In general, UTH larvae contained a higher level of total lipids and saturated FA and a lower percentage of unsaturated FA. BSFL fed control and fruit diets showed higher level of lipids and saturated FA, while the presence of meat in the diet increased the level of C20:4n6, C20:5n3 and C22:5n3. The C18:2e9t11 was also detected, especially in fat from larvae fed vegetable and fruit diets. The results confirmed that BSFL accumulate phytosterols in their lipid fraction. The sterol content between the two populations was significantly different, with a higher percentage in GRE larvae. Finally, BSFLs showed high plasticity in the lipid profile according to both the substrate and the population adopted. Nutritional characteristics of the BSFL may be shaped by modifying the substrate, in order to obtain tailored technological and feeding features.

Fatty acid profile of black soldier fly larvae reared on winery by-products

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The black soldier fly (BSF) is attracting interest for its ability to convert organic waste and by-products into high-value feeds. This study aimed to assess the effect of using, as rearing substrate, grape pomace (GP) from a red (Bequet, B) and a white (Moscato, M) variety on the fatty acid (FA) profile of BSF larvae (BSFL). The two GP were analysed for their proximate composition and phenolic fractions, while both GP and BSFL were analysed for their FA profile. The M-fed BSFL showed a higher total lipids (TL) content than the B-fed ones ($P < 0.05$). The contents of saturated ($P < 0.001$) and monounsaturated FA ($P < 0.05$) were higher, while those of branched-chain and polyunsaturated FA ($P < 0.001$) were lower in M-fed than B-fed BSFL. Interestingly, we detected the presence of conjugated linoleic acid (CLA) in GP (0.10 and 0.16 g/100 g TL in B and M, respectively), most probably as the consequence of the presence of lactic acid bacteria, which possess linoleate isomerase activity, involved in malolactic fermentation. In B-fed BSFL, we then observed a bioaccumulation of CLA up to 2.95 g/100 g TL, while levels remained considerably lower in M-fed BSFL (0.05 g/100 g TL). Several hypotheses can be postulated for such results, among which the presence of Stearoyl Co-A desaturase enzyme in BSFL and the presence of lactic acid bacteria in BSFL gut. In a circular economy perspective, winery by-products could be valorised within BSFL rearing media with the aim of obtaining BSFL enriched in FA of potential nutraceutical interest. (Supported by AgriTech National Research Center – PNRR CN 00000022).

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Efficiency of rearing yellow mealworms (*Tenebrio molitor* L.) on fibre-rich composted and mycoremediated residual streams

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Yellow mealworms (YM) have gained attention for their potential applications across various domains (food, feed, biotechnology, and waste management). They can transform organic waste, including food scraps and agricultural by-products into valuable products. This study aimed to assess the efficacy of rearing YM on low-value, high-fibre organic residual streams. Reed, grass, Japanese knotweed (JK) and nature straw were either ensiled, composted and inoculated with fungi or pasteurised and inoculated with fungi before being tested in two pilot experiments (50 YM/treatment) to identify optimal combinations for a subsequent large-scale experiment. In pilot 1, growth performance was evaluated across 44 combinations after 33 days of incubation. Composted JK exhibited better growth performance in terms of survivability (92%) and weight gain (5mg/larva) compared to the other treatments. In pilot 2, wheat bran (WB), corn starch and pea protein were incorporated to enhance the nutritional content of JK, while considering the effect of particle size through shredding. Thirteen treatments, including WB and ensiled JK as controls, were assessed in triplicates. The YM survival rate was high (>85%) across most treatments. However, larvae reared on JK:WB mixtures (10:90 and 30:70 ratios) displayed the highest weight gain (95 and 90 mg/larva respectively) and low FCR (~2 g:g). Conversely, the composted JK treatment without additives yielded lower larval weight gain (24 mg/larva). Thus, the inclusion of JK to WB may have likely improved the dietary structure but did not significantly augment nutrient provision.

New life for rice by-products through yellow mealworm bioconversion

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Tenebrio molitor (TM) is commonly reared on substrates with high dry matter (DM) content, thus including rice by-products (i.e., rice hull, rice husk, green grains, broken rice). However, information about the suitability of rice by-products as TM substrate are still limited. This study aims to evaluate the effects of rice by-products-based diets on growth and bioconversion efficiency of TM larvae. Five isonitrogenous (18% crude protein [DM]), isolipidic (3% ether extract [DM]) and isoenergetic (18 MJ/Kg [DM]) diets were tested (5 replicates/diet, 10000 3-week-old larvae/replicate, 0.26g/larva): C (control diet; feed waste, breeding waste, dry distillery stillage, coffee silvery film and hazelnut film), WR1 (10.5% of white rice by-products), WR2 (26.5% of white rice by-products), PR1 (17% of parboiled rice by-products), and PR2 (42.5% of parboiled rice by-products). Larval weight was recorded at the beginning of trial and every week. Specific growth rate (SGR), feed conversion ratio (FCR), survival, efficiency of conversion of digested food (ECD), and sieveability percentage (SP) were calculated at the end of larval growth (growth $\leq 50\%$). Data were analysed using SPSS software. Diet did not influence survival and development time ($P > 0.05$). However, all the rice by-products-based diets led to higher final larval weights and SGR than C group ($P < 0.05$). The lowest ECD was observed in WR1 and C larvae ($P < 0.001$), but both displayed the highest SP ($P < 0.001$). Lower FCR was also observed in WR2-fed larvae in comparison with C diet ($P < 0.05$). In conclusion, rice by-products are able to improve growth performance and bioconversion efficiency of TM larvae, with white rice being preferable in terms of SP. The research was supported by Fondazione Cariplo (newRIFF project).

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Optimizing the reproduction of *Tenebrio molitor*

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Numerous publications already indicate that *Tenebrio molitor* can be a great source of sustainable animal protein for both feed and food. However, in general these papers refer to environmental sustainability and not economic sustainability. The latter is often difficult due to a lack of specialized knowledge and equipment resulting in fairly high investment or operational costs. One of the ways to lower the costs is to improve the reproduction ensuring that less space and resources have to be allocated to this part. At the Insect Research Centre several experiments were performed in order to improve the reproductive output. In a first experiment the particle size of the feed of the adults was assessed. This parameter may affect the density of the feed and therefore the way beetles can borrow in the feed, layer thickness and 'air' gaps. It was observed the particle size of the feed had a major impact on the egg production with smaller particle size resulting in a higher output. In a second trial the effect of the size of the beetles (male and female) on the reproduction was assessed both on the willingness to mate and on the reproductive outcome. In a third trial the sex ratio and the possibility for multiple copulations was assessed. Preliminary results indicate that the beetle size has less of an influence than anticipated, while the sex ratio and multiple copulations did significantly alter the number of offspring. The combined results of these experiments will improve the reproductive output and decrease the space and time allocation.

Comparative larval growth of *Alphitobius diaperinus* populations on various substrates

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The lesser mealworm, *Alphitobius diaperinus* (Panzer), is a promising species for mass production due to its potential as a nutritious source for feed and food. Nonetheless, there is still plenty of room for improvement regarding its production. For instance, traditional livestock animal production typically involves the utilization of strains or breeds with favorable production characteristics for intensive farming. However, in the case of *A. diaperinus* the effect of the genetic material has not yet been thoroughly evaluated. To this end, the larval growth of two wild populations from Turkey and USA, one laboratory and one wild population from Greece, and two populations from commercial rearing facilities in the Netherlands was comparatively evaluated in laboratory trials. All populations were evaluated in flour of 100% wheat bran, rice hulls or sunflower seeds. The results indicated considerable differences among the populations with regard to their larval development, as well as to their feed conversion efficiency parameters. The study aims to highlight the significance of selecting *A. diaperinus* populations with specific traits to improve the overall efficiency of the farming systems. The research work was partially supported by the Hellenic Foundation for Research and Innovation (HFRI) under the 4th Call for HFRI PhD Fellowships (Fellowship Number: 11297) and by the COST Action <Improved Knowledge Transfer for Sustainable Insect Breeding (Insect-IMP), CA22140>, supported by COST (European Cooperation in Science and Technology).

Session 11

Theatre 13

Black soldier fly: progress and challenges

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Most insect-derived products are used as petfood and the rest as aquafeed, and poultry and pig feed. This to replace fishmeal or soybean meal, the use of which is questioned because of sustainability reasons. Plant proteins are often used but there are nutritional problems. The black soldier fly (BSF) can be reared on a wide range of organic side streams. Ninety percent of all BSF publications are from the last six years. BSF larvae naturally produce a diversity of antimicrobial peptides to protect themselves from the environment full of microorganisms in which they live. These peptides exert a positive impact on animal health and can be used as potential antibiotic replacements. In mass rearing, BSF's immune system can be influenced through the microbiome. Up till now hardly any disease has been reported for BSF. Companies select BSF strains to convert certain waste streams efficiently. Abiotic factors can influence the protein and fat content of BSF, while waste streams can influence the fat but not the protein content. Depending on the aquaculture species, up to 100% full fat or defatted BSF meal can replace fishmeal. In poultry and pig feeds, 10% of plant protein sources can be replaced. Live BSF larvae also promote the natural foraging behavior of poultry (reduced feather picking) and pigs (reduced tail biting). In pet food, inclusion of BSF meal may have health hypoallergenic effects. In the European Union, regulatory frameworks for insect-derived products are slowly becoming more conducive. BSF fats can be used as biodiesel or fuel, in cosmetics (skincare products), and as surfactants, and protein for making bioplastics. BSF is increasingly used for bioremediation purposes. Left-over substrates (frass and exuvia) can be used as fertilizer, crop protection agent, and for drought control. Nanoplastics can move up the food chain, with BSF larvae as intermediate. The price of BSF meal is still too expensive but is expected to go down. Innovations are being expected in automation of production techniques, optimization of bioconversion (efficient interaction between microbes in the insect gut and feed substrate), disease management, selection of effective strains, food safety issues and processing.

Assessing the optimal carbon-to-nitrogen ratio to improve black soldier fly larvae production for future use as animal feed

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The global livestock production industry is expanding as the world population increases, resulting in a high demand for animal feed. In particular, protein sources such as soybean meal and fish meal are becoming increasingly scarce and costly annually. Therefore, studies to find a sustainable alternative feed source to reduce reliance on limited resources, such as black soldier fly larvae (BSFL) are of great interest. However, BSFL still exhibit several limitations that require additional research for a comprehensive understanding, particularly in optimizing the essential nutrients for their growth. The aim of this study was to investigate the optimal carbon-to-nitrogen ratio (C/N ratio) for BSFL. Cassava pulp and layer manure were selected as carbon and nitrogen sources, respectively. A batch of 5-day-old BSFL was divided into eight groups, each comprising six replicates. The experimental diets are as follows, Group 1 : control group (commercial chicken feed, 21% CP), while groups 2 through 8 were subjected to varying C/N ratios of 10:1, 15:1, 20:1, 25:1, 30:1, 35:1 and 40:1, respectively. The BSFL were reared until the prepupae stage, with observations initiated from the appearance of the first pupae in each treatment group, followed by harvesting the next day. The results indicated that feeding the BSFL at a 15:1 of C/N ratio revealed the most optimal productive performance. Moreover, it showed positive results in waste-to-biomass conversion rate and waste reduction rate ($P < 0.05$). This suggests that the optimal ratio of C/N ratio for BSFL growth is at 15:1. Future research should focus on investigations into the use of 15:1 C/N ratio with other alternative materials.

Promoting insect farming sustainability with the utilization of oilseed presscakes as nutrient-rich feed for *Tenebrio molitor* larvae

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Coupled with the valorization of agricultural byproducts, insect farming aims to fulfill the principles of circular economy. In this context, recent research has focused on the utilization of a variety of agricultural byproducts as insect feed, in order to close the loop in the food chain by repurposing agricultural waste. However, the potential of oilseed presscakes, the byproduct of oilseed processing, has not been thoroughly investigated yet. This study investigates the efficacy of utilizing oilseed presscakes as feeding substrates for larvae of the yellow mealworm, *Tenebrio molitor*. The tested presscakes derived from the production of pumpkin, sunflower, cannabis, linseed, thistle, walnut, almond, and sesame and all had a high protein, lipid and energy content. For the bioassay, 50 early-instar larvae were introduced in vials (7.5 cm diameter) and fed with each of the presscakes, while agar served as moisture source. Larval survival and growth were evaluated until the emergence of the first pupa. The feed conversion ratio (FCR) and the total development time were also determined. The results demonstrate the potential of oilseed presscakes as valuable feeding substrates that can provide balanced nutrition for *T. molitor* larvae. Furthermore, this research underscores the sustainability of insect-based protein production systems within the circular economy framework. Financial support for this research has been provided by PRIMA, a program supported by the European Union, under grant agreement No 2231, project CIPROMED (PRIMA Call 2022 Section 1 Agri-food IA).

Performance and bioconversion of black soldier fly across different content of fiber and protein in the substrates

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This study investigates the bioconversion efficiency and nutrient accumulation in black soldier fly (BSF; *Hermetia illucens*), focusing on the effects of feeding on two levels of fiber (low, 9.2; high 18.5) and three levels of crude protein (low 13.5%, medium 20%, high 26.5%) within a 3×2 factorial arrangement. Results showed that BSF reared on high fiber showed higher survival rate, substrate consumption, and higher lipid yield, as well as lower bioconversion, protein conversion, and protein yield. The weight, length, protein yield, and lipid yield were greater in medium and high protein than low protein. The analysis did not show significant differences in dry matter, ether extract, ash, and chitin content. The content of gross energy in harvested BSF was higher in high fiber diets. The content of CP was higher in BSF fed high or medium protein levels compared with the low protein. Amino acid profiling revealed increased concentrations of isoleucine, leucine, phenylalanine, threonine, tryptophan, alanine, aspartate, glycine, proline, serine, and tyrosine in high fiber treatment. Isoleucine, leucine, tryptophan, alanine, glycine, proline, tyrosine was higher in BSF fed high or low protein levels compared with medium. Fatty acid analysis indicated that the ratio of C12:0, C14:0, C15:0, and C20:1 was higher in BSFs fed higher fiber treatment compared with low fiber treatment, while C18:4 was lower in the low fiber treatment. There was no significant difference in the fatty acid analysis in the protein level treatment. In conclusion, BSF fed higher fiber and protein levels emerged as a superior substrate for enhancing performance and the bioconversion potential of black soldier fly.

Session 11

Poster 17

Nutritional profiling and efficiency of black soldier fly bioconversion across different substrates and developmental stages

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This study investigates the bioconversion efficiency and nutrient accumulation in black soldier fly (BSF; *Hermetia illucens*), focusing on the effects of feeding on two different substrates (tofu by-products and food waste) and harvesting at two developmental stages (larvae and prepupae) within a 2×2 factorial arrangement. Results showed that BSF reared on tofu by-products significantly outperformed those on food waste in terms of weight gain. Additionally, harvesting at the prepupae stage resulted in enhanced weight, length, width, and substrate utilization, although larvae harvested BSF exhibited higher survival rates. In terms of bioconversion metrics, BSF fed on tofu by-products demonstrated greater protein conversion efficiency, protein and lipid yield. However, lipid conversion was greater in BSF consuming food waste. The conversion efficiencies and yields for protein and lipids were higher when BSF were harvested at the prepupae stage, whereas larval stage harvesting showed greater overall bioconversion rates. Harvesting at the prepupae stage led to higher levels of crude protein, ether extract, and chitin. Amino acid profiling revealed increased concentrations of ile, leu, trp, ala, and pro in BSF fed tofu by-products, with greater thr, trp, ala, ser, and tyr in prepupae-stage. Fatty acid analysis indicated that C10:0, C12:0, C14:0, C14:1, C18:3n3, and C20:1 was higher in BSF consuming tofu by-products. Additionally, the saturated fatty acids, C12:0, C15:0, C17:0, C18:1 were more abundant in BSF harvested at the prepupae stage. The BSF meal derived from tofu by-products showed higher gross energy, and meals from prepupae-stage BSF had enhanced gross energy, crude protein, and chitin content, along with a distinct fatty acid profile favoring C12:0, C18:3n3, C22:6n3, and higher levels of saturated fatty acids. The MDA content was significantly lower in BSF meals from prepupae after 14 and 28 days of storage, suggesting better lipid stability. In conclusion, tofu by-products emerged for enhancing protein and lipid yields, while the prepupae stage offered advantages in nutrient density and shelf stability of the harvested biomass.

Fatty acid composition of *Hermetia illucens* larvae grown on different expired wet pet-food

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The present study compared the effects of four different discarded pet foods (for cats) on *Hermetia illucens* fatty acid compositions of neutral (NL) and polar lipids (PL). We used: 1) broiler feed as a control diet (CTRL); 2) salmon mousse (SLM); 3) chicken mousse (CHK); 4) beef mousse (BF). Each diet was put into 9 plastic containers (n=9); each one had 2.5 kg of substrate and 1,200 five-day-old larvae. At 22 days old, larvae were separated from the frass, fasted for 24 hours, frozen, and then chemically analyzed. Total lipids (TL) were extracted, and then NL and PL were separated by solid-phase extraction. Each lipid fraction was methylated to obtain the fatty acid (FA) profile by gas-chromatography. A one-way ANOVA was run to highlight statistical differences. Results showed that SLM larvae had the highest TL content (26.17 g/100 g of dry matter). Saturated fatty acids (SFAs) prevailed over other FA categories, with a substrate effect observed. Specifically, BF, CHK, and SLM larvae had a lower content of SFAs and a higher content of monounsaturated FA (MUFAs) compared to the CTRL larvae. SFAs were preferentially stored in NL, while MUFAs were stored in PL. SLM pet food slightly enriched the larvae in n-3 polyunsaturated FAs, which were mainly stored in PL. In conclusion, *Hermetia illucens* can represent a promising avenue to avoid incineration or landfill disposal of waste, but emphasis on the accumulation site of essential fatty acids should be considered.

Effects of herbs dietary inclusion on growth performance and nutrient composition of *Tenebrio molitor* larvae

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Mealworm larvae (MW, *Tenebrio molitor*) are able to convert grain byproducts (i.e. bakery waste) into high-quality biomass, which can be processed into animal feed. Since moderate inclusion of several herbs may occur in some bakery waste material, this study examined the effects of herbs dietary inclusion on growth performance and nutrient composition of *Tenebrio molitor* larvae. The MW larvae were reared on wheat bran (control diet), or supplemented with four different herbs, namely, basil, oregano, rosemary and thyme supplemented in two concentrations (0.125% and 2% dry matter). All experiments were conducted under light condition, to counteract the meal moths, at 27°C with 60% relative humidity for five weeks. Growing performance parameters were recorded during the trial and nutritional composition was analysed on resulting larvae. The growth performance showed no differences between groups. Considering the DM content, no differences were observed among groups. In the case of protein content, greater inclusion of herbs resulted in lower protein levels. Regarding the fat level, only the inclusion of oregano led to lower content compared to the control group. Concluding the dietary inclusion in MW larvae of selected herbs up to 2% did not affect growth performance. However, some minor detrimental effects have been observed in the case of fat content in larvae receiving oregano. Further studies are needed for understanding the observed effects.

Prospects of breeding programs for small ruminant breeds in the Mediterranean region

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Sheep and goat farming is widespread throughout the Mediterranean region with systems that vary from intensive with improved breeds to extensive with local breeds well adapted to the environment in areas not suited to intensive agriculture otherwise destined for depopulation. The attempts made since the 1990s to develop modern breeding schemes have achieved significant results for few dairy breeds that have spread in less favorite areas, as purebred or crossbreeds, not always with appreciable outcomes. For most local breeds, the difficulties of overcoming the laboriousness of selection tools (phenotype recording, artificial insemination, single sire mating) have severely limited their effectiveness. Over the last decade, the need to organize the genetic management of local breeds has emerged with even greater force, not only to improve their economic sustainability and therefore mitigate the risk of extinction of some of them, but also to preserve their genetic potential for adaptation to climate change. At the same time, the advent of new genomic technologies and the constant reduction in the cost of their application makes a change in the traditional “large scale recording-artificial insemination” paradigm possible. An overview of the state of the art of breeding schemes is provided focusing on the potential of new genomic technologies for both genetic improvement and conservation of genetic variability. Particular focus is given on the development of shared strategies and methodological approaches throughout the Mediterranean region to respond to common environmental, economic and social challenges.

Session 12

Theatre 2

Derivation of economic weights for sheep breeding goal traits considering production system characteristics

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The implementation of modern sheep breeding programs implies the development of new breeding goals considering novel functional traits. Theoretically, breeding goals should be specific for the respective populations and production systems. The aim of this study was to derive economic weights for sheep breeding goal traits considering characteristics of the herds, environments and social-ecological effects. Herd-environmental characterisations considered 13 traits from 25 surveyed farms. The application of different clustering methods (agglomerative hierarchical, partition around medoids and fuzzy) allocated the herds to 3 main groups. Major group differences were due to the effects of the farm type, production focus and herd size. The ongoing contingent valuations based on the “willingness to pay” for breeding goal traits indicated the importance of the traits maternity and parasite resistance in all clusters, with weighting percentages of 8,06% to 13,5%, respectively. The traits maternity and suitability for landscaping were especially important in cluster 3, which represents a large number of farms neglecting commercial meat production. Validations for the derivation of economic weights were performed through bio-economic modelling approaches, and applying the software “EcoWeight”. With the additional distinction between direct and maternal effects, the highest relative economic importance was derived for survival rate (direct component 24%, maternal component 31%). Sensitivity analyses highlighted the importance of meat prices on economic weights, especially for survival rate. In contrast to the bio-economic approach favoring the weighting of production traits, contingent valuations emphasized functionality.

Intra-island patterns of genetic divergence in Cyprus Chios sheep: minimizing genetic background noise to enable more precise genetic improvement

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The effects of climate change, the ongoing global economic crisis, and the rapidly shifting geopolitics, particularly in Eastern Europe, indicate the need for food security and hence the Primary Production Sector's importance. Being on the southeastern border of Europe, the island of Cyprus experiences limited feed production mainly due to unfavorable environmental conditions, while at the same time facing a constant increasing demand for sheep and goat milk. This requirement is a result of both the expanding local population and the recent registration of Halloumi cheese as a product with Protected Designation of Origin, for which, Cyprus is bound by EU regulation, to increase the product's goat/sheep composition to 51% by 2029. Towards this objective, the Agricultural Research Institute, coordinates a four-year project to genetically improve the local Cyprus Chios sheep in terms of total milk yield and quality characteristics. Exploiting publicly available genome-wide SNP data from circum-Mediterranean sheep breeds, a 100-subset of SNPs was proven to effectively identify individuals belonging to the local breed. Aiming to minimize the genetic background noise on current improvement efforts on the focal Chios breed, the same tool was used to assess the within breed genetic diversity and admixture profiles, including >5000 samples from 24 farms in our downstream analyses. Generated results indicate unexpectedly high genetic divergence between examined farms, highlighting the importance of estimating the genetic profile of each population before targeted reproduction. At the same time, gene flow routes are in line with geographic proximity and known exchanges of males were revealed. Research findings are anticipated to promote more precise genetic improvement and facilitate subsequent GWAS analyses and improve farm management practices, such as control of inbreeding.

Genomic analysis of dominance and inbreeding effects on milk production traits in Pag sheep

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Local breeds are reservoirs of genetic diversity and have phenotypic characteristics that could be an important resource for meeting future challenges to livestock systems. For example, Pag sheep are a local Croatian breed adapted to a Mediterranean island with a marginal grazing system, raised for milk (mainly used for cheese) and lamb production. Sustainable selection programmes for small local breeds like Pag sheep, may need to consider the effects of dominance and inbreeding. Our aim was to estimate additive and dominance genetic variances and genomic inbreeding for milk traits (milk, fat and protein yields and somatic cell score). We had genotype data (50K SNPs) for 2134 animals, while 1744 ewes had milk records. We detected runs of homozygosity (ROH) with plink 1.9 to estimate genomic inbreeding (FROH). The four single-trait models with additive and dominance effects, including FROH as a covariate to account for directional dominance, were fitted both via the Bayesian method and REML as implemented in BLUPF90 software. Our results revealed stable additive variance across different models. In contrast, dominance variance varied across models, and was significantly impacted by whether or not the FROH covariate was in the model. Dominance variance accounted from 10-30% of genetic variance across models and traits. In conclusion, SNPs allow us to estimate dominance variance and inbreeding in small populations, which can contribute to the knowledge needed to develop sustainable genomic selection programmes in such populations.

Evaluation of crossbreeding potential in heavy suckling lambs obtained by crossbreeding Sarda dairy ewes with Dorper rams

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The aim of this study was to evaluate the growth performance, carcass characteristics and meat quality of lambs obtained by crossbreeding Sarda dairy ewes with Dorper rams. Twenty single-born Dorper x Sarda (DS) lambs (9 males, 11 females) and 19 single-born Sarda (S) lambs (10 males, 9 females) were used. Lambs were fed exclusively with maternal milk until slaughtering at 11 weeks of age and with a body weight (BW) of 20 ± 0.28 kg of (mean \pm SD). During the trial, BW was measured twice a month for estimating average daily gain (ADG). At slaughter, carcass measurements, pH and colour traits were recorded. Proximate composition, fatty acid (FA) profile, cooking and drip losses, and consumers perception were evaluated. Almost all data were analysed by a PROC GLM procedure to test the differences between the genetic type, sex, and their interactions. The ADG did not differ between S and DS lambs and between males and females. The S carcasses had a higher fat content and rib fat thickness compared to DS lambs. Intramuscular fat of DS lambs had a higher content of 22:5n-3, 22:6n-3, branched-chain FA, and odd- and branched chain FA. Both DS and S lamb meats presented no distinguishable quality characteristics from consumers perception. In conclusion, crosses between dairy Sarda breed, and the meat Dorper breed seems a valuable opportunity to improve meat value and accessibility, although the commercial viability of producing this new type of lamb remains to be determined. Acknowledgements: research funded by PROBOVIS project (PSR Sardegna 2014–2020; sottomisura 16.2) and RESTART-UNINUORO project (Regione Autonoma della Sardegna; FSC 2014–2020).

Using genotypes from Australia's numerically smaller sheep breeds to expand the genomic reference population

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Genetic evaluation of animals with different population ancestry needs to deal with differences in their founder populations or breeds. To accommodate these differences allele frequencies from different genetic groups can be used to correct the genomic relationship matrix to reduce breed and group structure. The majority of the Australian sheep population is comprised of animals originating from 11 major breeds. However, smaller breeds are also used to create crossbred animals for different breeding objectives. To properly include these animals into a multi-breed genetic evaluation, the allele frequencies of smaller and often missing breeds need to be included into the allele frequencies of the wider population. This work investigated the use of genotypes from smaller breeds and crossbred populations that are currently not included in the Australian national genetic evaluation (LAMBPLAN) because reference populations for imputation and inclusion in breed adjusted genomic relationship matrices have not been established. Genotypes from 50,999 animals originating from fifteen numerically smaller meat, wool and maternal sheep breeds were combined in a genetic analysis to investigate population structure and diversity within and across breeds and define breed composition. Results are expected to increase the number of animals included in the genetic evaluation and improve prediction accuracy in Australian crossbred populations.

Phenotypic and genetic parameters of lactation persistency and milk yield of patch-faced Maritza sheep breed
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The aims of this study were to estimate the phenotypic and genetic parameters of lactation persistency (LP) and milk yield (MY) in sheep. For this purpose, a database of 643 lactation records for 351 ewes of the Patch-faced Maritza sheep breed for the period 2018-2023 was used. Pedigree file included 586 animals, of which 351 ewes had records, as daughters of 35 rams. Each lactation record per ewe included 5 test day milk yield (TDMY) records during the milking period (MP). Lactation persistency was calculated as the ratio of average TDMY to maximum TDMY over the course of MP for each ewe, expressed as %. The average suckling period (SP) was 70.79 days (± 16.61), average MY per 150 daily MP was 115.57 L (± 34.43). Average daily MY was 770.49 mL (± 229.25) and average maximum TDMY – 1271.10 mL (± 447.63). The average LP was 62.43 % (± 9.70). Variance components of LP and MY were estimated by using two types of animal models: A single trait repeatability animal model (AM) for each trait and a simultaneous two trait repeatability AM. All analyses were performed in an R software environment. The SP was included as a regression and the age of the ewe was included as a fixed effect. Additionally, flock and farming year were concatenated as common fixed effect in the models. In the simple repeatability AM, the heritability (h^2) of LP was 0.112 ± 0.020 and the repeatability (rw) was 0.141 ± 0.040 . The heritability of MY was 0.220 ± 0.025 and rw was 0.315 ± 0.060 . Regression of SP on LP was 0.0022 but for MY -0.360. For the two traits, a simultaneous two trait repeatability AM, the h^2 of the LP was 0.092 ± 0.018 and the rw was 0.132 ± 0.038 and h^2 of MY was 0.208 ± 0.024 and rw was 0.276 ± 0.057 . Phenotypic and genetic correlations between LP and MY were respectively -0.047 and 0.038. Regression of SP on LP was 0.0022 but on MY was -0.357. The lack of genetic correlation between LP and MY indicates that selection for high milk yields may not result in high persistency.

Session 12

Theatre 8

Enhancing breeding programs for local breeds in challenging environments: insights and innovations

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In harsh environments, the value of genetic advancements may be overshadowed by environmental fluctuations, making it more difficult to discern incremental improvements and harder to justify investing in breeding programs. In such scenarios, expanding performance recording becomes imperative to monitor the impact of management interventions, mitigate the risks associated with inbreeding, and provide the supply chain with useful information. Initiatives such as the sheep and goat community-based breeding programs in Ethiopia and India (Mesha project) show that flexible but robust data tools play a pivotal role in supporting breeding efforts in challenging socio-technical and environmentally harsh contexts. In scenarios where small ruminants are essential assets for smallholder farmers, these animals offer a means to alleviate poverty and provide manageable income streams. Production efficiency gains in these small ruminant populations will also reduce the greenhouse gas emissions intensity associated with the products, and enhance the social and ecosystem services they provide. Novel sustainable approaches that extend beyond the traditional national systems focused on breed purity, can deliver multifaceted benefits beyond genetic improvement. The potential use of composite or cross-breeds is a promising avenue for enhancing productivity and resilience. By addressing these challenges and leveraging innovative strategies, breeding programs can be optimized in harsh environments, fostering resilience and sustainable development in farming communities worldwide. This approach can promote sustainable growth and development in smallholder farming communities, and beyond.

Genomic insights into Milk Coagulation Properties and Curd Firmness in Pag Island Sheep: a Genome-Wide Association Study

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Efficient cheese production relies on optimal milk coagulation, which is a key factor for cheese quality. In this study, focussing on the largest dairy sheep population in Croatia, the Pag Island Sheep, a Genome-Wide Association Study (GWAS) was conducted to identify polymorphic variants associated with traditional milk coagulation properties (MCP) and new curd firmness over time (CFT) model parameters. These parameters are obtained from a mathematical model that, differently from the traditional MCP, uses all the information yielded by the instrument. Our analyses were based on 1173 milk samples, all analysed with the Formagraph instrument, and genetic data from 291 animals genotyped with the GeneSeek® Genomic Profiler™ Ovine 50k using the BLUPF90 family programme and the GRAMMAR method. While considerable influence of the polygenic component was observed, our analyses identified specific single nucleotide polymorphisms (SNPs) that contribute significantly to MCP variation. This is the first GWAS in “Western Balkan sheep breeds”, providing crucial insights for the genetic improvement of Pag Island Sheep to increase the efficiency of cheese production. Funding: “Potential of microencapsulation in cheese production” (KK.01.1.1.04.0058), Operational Programme Competitiveness and Cohesion 2014–2020 and “Pheno-Geno-IP-2022-10-6914” funded by Croatian Science Foundation.

Session 12

Theatre 10

Estimation of genetic parameters for fertility and prolificacy in the Lacaune meat sheep population carrying a hyperprolific gene

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Numerical productivity, a key economic criterion in meat sheep farming, is influenced by ewe fertility (FERT) and prolificacy (PROL). This study looked at these two production traits and two of their main factors of variation: the method of reproduction (on induced oestrus, IO, and on the first natural return oestrus, NRO) and the genotype of the FecL hyperprolific gene. The dataset covered the reproductive records of 20,770 Lacaune meat ewes from the Ovi-Test breeding company, 90% and 58% of which had a recorded sire and FecL gene genotyping, respectively. Between 2010 and 2020, 68,652 matings resulting in 49,248 lambings were analysed. The aim of this study was to first confirm the effect of genotype on prolificacy in our dataset. Subsequently, we assessed the effect of genotype on fertility and estimated the genetic parameters of FERT and PROL traits. The genetic evaluations were performed using Asreml software based on a BLUP animal models: a two-trait model PROL_IO and PROL_NRO and two single-trait models for FERT_IO and FERT_NRO. The estimated effect of FecL gene was similar to previous study (+ 0.5 lamb per litter for heterozygous carriers compared to non-carriers) but had no significant effect on fertility. The estimations of animal genetic variances for the two traits on IO or NRO were low but in line with the literature: for prolificacy, $\sigma^2_{a_IO} = 0.046$ and $\sigma^2_{a_NRO} = 0.043$; for fertility, $\sigma^2_{a_IO} = 0.007$ and $\sigma^2_{a_NRO} = 0.007$. The heritabilities of PROL_IO and PROL_NRO were 0.06 and 0.09 respectively, with a genetic correlation of 0.95. The heritabilities of FERT_IO and FERT_NRO were 0.04 and 0.03 respectively.

Early-life trait effects on the reproductive performance of Cyprus Damascus goats

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Goat reproductive performance, as assessed by traits such as pregnancy to first breeding, age at first kidding, kidding interval, litter size, litter survival as well as kids weaned by the dam, is a major determinant of productivity and economic viability of caprine rearing systems. A number of studies have previously attempted to define both genetic and non-genetic factors associated with reproductive traits in various goat breeds around the world. However, a lack of goat breed-specific information and kid-rearing benchmarks may limit farmers' ability to select the best animals to keep as breeding stock as well as to control aspects that affect the herd's overall fertility, fecundity, prolificacy, and future farm productivity. We performed a retrospective analysis based on individual goat data that are systematically recorded in the Cyprus Damascus goat herd maintained in the Agricultural Research Institute, Cyprus, to test the potential effect of early-life characteristics (birth weight, birth type, kidding season, kidding year, pre- and post-weaning weight and growth rates, and the rearing system used before weaning) on reproductive performance component traits of Cyprus Damascus goats. The study cohort included 34 mating seasons covering a period of 17 years (2005-2022). Descriptive statistics followed by multivariate analyses were conducted to evaluate intrinsic and extrinsic factors that could be used to predict 1) pregnancy to first breeding, 2) age at first kidding, 3) kidding interval, 4) litter size at birth, 5) litter size at weaning and 6) survival rates of kids at birth and at weaning. Heritability estimates were determined based on available pedigree information. The findings would not only aid in the selection of breeding goats with desirable characteristics, but they would also be useful in future genome-wide association studies to investigate genetic effects and theories based on these discoveries.

Session 12

Poster 12

Benefit of including female genotypes in the reference population of French Lacaune dairy sheep, a simulation study

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In the French Lacaune dairy sheep breeding program, genomic selection is presently based solely on a male reference population. The benefit of including female genotypes is questionable. This study evaluates, using a deterministic gene flow model, the increase in genetic gain and economic implications of enhancing the accuracy of genomic Estimated Breeding Values (GEBV) through genotyping of ewes. Over a 20-year period, the genotyping of females has not proven economically viable at the nucleus level, regardless of the gain in genomic EBV accuracy. The consideration of ewe lambs as elite dam candidates consistently improved outcomes due to a reduction in the generation interval. However, profitability at the nucleus level remained very low or even non-existent depending on the genotyping cost considered. Extending the cost-effectiveness analysis to the broader Lacaune population (including nucleus and commercial flocks) yielded superior economic results, regardless of the magnitude of genomic EBV accuracy gains. Nevertheless, profitability remained unattainable at moderate accuracy enhancements. Given the logistical challenges and limited profitability, widespread genotyping of ewes seems inappropriate in the current situation. A necessary reduction in genotyping costs, coupled with co-products such as parentage verification and management of specific alleles, is deemed necessary to make large-scale ewe genotyping economically viable.

Genetic diversity of related sheep breeds Sarda and Pag

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Genetic diversity of sheep is endangered, mostly by the replacement of local breeds with commercial breeds. However, local breeds are adapted to unique environments, rustic, resilient, a reservoir of genetic diversity, and could help with meeting challenges due to climate change and market demand fluctuations. Two examples of local island sheep in the Mediterranean region are the Sardinian White (Sarda) and Pag breeds. Sarda sheep are widely found in Italy, while Pag sheep are a Croatian breed that likely had historical introgression of Sarda. Our aim was to study the genetic structure of these Sarda and Pag breeds to highlight differences and common genomic regions, that might indicate environmental adaptations. Using 50K SNP data from 825 Sarda and 2683 Pag, runs of homozygosity (ROH) were detected using detectRUNS in R, and F_{ST} analysis was done using PLINK. Sarda ($F_{ROH}=0.07\pm 0.04$) had more inbreeding than Pag ($F_{ROH}=0.04\pm 0.05$), probably due to lower selection pressure on the latter. A total of five ROH segments were shared between the two breeds. We identified 367 SNPs with a $F_{ST} > 3$ SD from the chromosomal mean or that had a F_{ST} in the top 99th percentile. The analysis of genes mapped on shared ROH segments or close to significant SNPs could help understand the history of these two breeds and how past introgression contributed to today's Pag sheep.

Session 12

Poster 14

Using cryptorchid rams to induce a ram effect in a flock of ewes and successful insemination upon natural heat

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The use of hormones for animal synchronization is facing increasing restrictions. Using aprons on fertile rams or vasectomizing them is challenging from an animal welfare standpoint. However, artificial insemination is still an important tool for genetic improvement and creating genetic links among flocks. Cryptorchid rams are characterized by undescended testicles, which cause azoospermia while maintaining near normal levels of testosterone. Moreover, these rams were shown to successfully detect females in heat. The objective of this study is to assess the capacity of these rams to induce a ram effect on an ewe flock, detect those that go in heat and check the success of artificial insemination after natural heat. Four cryptorchid Sarda rams were introduced to 31 Sarda ewes. At day 14, rams were removed, and one was reintroduced 4 times a day from day 16 until day 25 to detect ewes in heat. Out of 31 ewes, 22 (70.97%) responded to the ram effect with subsequent heat signs. Eleven ewes were arbitrarily chosen and inseminated 24 hours after heat detection with chilled semen (400 million sperm/dose, conserved at 4°C and used within 24 hours). Out of these ewes, 5 gave birth (45.45% fertility rate). These results show that cryptorchid rams can induce a ram effect, detect ewes in heat and are a safe and reliable tool to induce a natural heat upon which artificial insemination can be performed. This natural technique can substitute the use of hormonal treatments, thus satisfying consumer welfare demands. They can also replace the use of vasectomized rams or the aprons on fertile rams leading to an improved animal welfare.

Quantitative-genetic characterization of wool quality traits in German Merino

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The estimated breeding value (EBV) for wool in German sheep breeding programs is based on a subjective assessment of the fleece. There is no possibility to select for improved fiber quality yet. As global wool prices have gone down, prices for German wool went down close to zero. Criticism from the industry is that the quality is not defined and the fiber diameter is too large and fluctuating. We are now carrying out quantitative-genetic analyses of wool quality traits, i.e. traits derived from fiber measures, in German Merino sheep in order to create the basis for EBVs and a breeding program to improve wool quality. Better quality should improve the use and thus the producer prices of German wool as a regional product on the domestic market. In this study, wool samples (shoulder, back, loin) were taken from more than 2,000 breeding dams and sires of German Merino sheep in southern Germany (1,680 animals were also genotyped using 50K-SNP chips). The phenotypes were recorded via optical measurements using an ofda2000, recognized by the International Wool and Textile Organisation. Descriptive statistic procedures were then conducted in order to get an overview on the phenotypes, which have never been observed in a comparably large sample in this breed. Population-specific parameters and heritability were estimated for several wool quality traits by means of mixed linear models. In line with the literature, the study shows the potential of breeding for improved wool quality. However, further research is required before an implementation in the German Merino breeding program.

Evaluation of milk quantity and quality traits using BLUP models and longitudinal lactation records of a Cyprus Damascus Goat population

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Small ruminant breeding is of great financial and environmental importance around the world. The Damascus goat is of Syrian origin and was imported in Cyprus in early 20th century in order to increase milk and meat yield and has since been genetically improved and become a local breed. The Agricultural Research Institute (ARI) is the core research center in Cyprus for ruminant genetic improvement. Research interest of the ARI Animal Production unit focuses on the genetic improvement of the Cyprus sheep and goat populations under the AGRICYGEN project and a connected measure within the Cyprus Recovery and Resilience Plan (RRP) (NextGenerationEU). Currently, selection of high producing animals in Cyprus is achieved by combining pedigree information with selection indices for the evaluation of phenotypes on growth rate in young animals and milk production capacity of their ancestors. Our recent pilot study implementing BLUP models on lactation records collected between 2008-2022 from 1104 ARI Cyprus Damascus goats, enabled the estimation of milk trait breeding values and the evaluation of current methods used in the selection scheme. The study revealed high heritability and genetic trends for annual milk yield, fat and protein content for the 1st lactation of Cyprus Damascus goat. Therefore, within the present study, analysis will expand on all lactation periods using ASReml package on R studio to evaluate our goat population for milk performance during their lifespan. Lifelong production is crucial and needs to be considered for successful animal breeding. In the future, the study will be further expanded to pursue genome-wide association studies (GWAS) for milk traits, including data from private Cyprus Damascus goat flocks, as part of an Aid Scheme under the Cyprus Resilience and Recovery Plan, funded by the European Commission.

Adoption of computer vision algorithms to monitor respiratory rates in dairy cattle

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The respiratory rate in dairy cows is a crucial parameter to be monitored, given its potential use as input data to detect early disease and track heat stress and reproductive performance from a herd-level perspective. Computer Vision (CV) is a multidisciplinary field of study that enables computers to extract information by analyzing images and videos in various scenarios, by developing algorithms typically included in the artificial intelligence domain. The objective of our investigation is to adopt Spatiotemporal Convolution techniques to monitor the respiratory rates of cows during milking in a herringbone parlour. Video files were recorded over six months, fragmented into 30-second segments, and subsequently annotated manually, counting the number of respirations and identifying animal movement. This resulted in 695 annotated videos used to train the CV model. The modified version of the video ResNetR(2+1)D architecture with R(2+1)D convolutional blocks achieved an accuracy of 93.2% and a Mean Absolute Percentage Error (MAPE) of 21.8% for frame sequence classification and respiratory rate regression, respectively, on our validation dataset. Our results are promising, encouraging further investigation, using a larger dataset comprised of animals from different herds across a whole year.

Session 13

Theatre 2

A novel method to differentiate and profile ruminal vs. breath volatile organic compounds for metabolic assessment in dairy cows

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Frequent eructation in ruminants blends exhaled ruminal and breath volatile organic compounds (VOC), limiting breathomics applicability for metabolic assessments in dairy cows. The aim of this study was to develop a protocol (SOP) to differentiate the origin of exhaled VOC. Samples from 30 multiparous lactating Holstein cows were analyzed. Exhaled VOC in breath (Br; blood borne metabolites) and exhalome (Ex; a mixture of ruminal eructation and Br) were sampled using a GreenFeed (GF). Methane (CH₄) levels and eructation events served as the marker to distinguish Br vs. Ex. Samples were analyzed using gas chromatography (GC, n=138) and secondary electrospray ionization mass spectrometry (n=738). Logarithmic transformation on Br over Ex mean ratio and fold change (FC) were calculated to differentiate metabolite's origin. Our results revealed CH₄ concentration 76% higher in Ex compared to Br (556 vs. 137 ppm, respectively). Based on our findings, a SOP was proposed using CH₄ GF readings >250 ppm for Ex and a threshold of <150 ppm CH₄ for Br sampling. Implementing this SOP, we identified ruminal VFAs in the Ex (P < 0.01) with a differentiation of FC > 0.5. TCA cycle precursors were identified in Br (P < 0.01) with a differentiation of FC > 0.2. Our study established a method to differentiate Br and provided evidence of the potential to implement breathomics as a non-invasive tool for conducting metabolic assessment in ruminant research.

Development of an HPLC method for the quantification of allantoin, creatinine and uric acid in Holstein cows' urine

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Many phenotypes related to heat stress can be identified in cows' urine, including compounds from protein metabolism, such as urea, creatinine, uric acid and allantoin. This study aimed to qualitatively and quantitatively determine allantoin, creatinine, and uric acid in Holstein cows' urine through reverse-phase HPLC. A total of 21 commercial herds located in Northern Italy were visited, and 1,275 individual urine and milk samples of Holstein Friesian cows were collected during morning milking. Cows produced on average 35.20 kg/d of milk, with 3.75% of fat and 3.40% of protein. Aliquots of 1 mL of each urine sample were centrifuged at 13,000xg for 15 min at room temperature. The supernatant was diluted in the proportion 1:10 with ultrapure water and filtered using a 0.22 µm syringe filter. Analysis of allantoin, creatinine, and uric acid was carried out using HPLC station (Agilent Technologies, Santa Clara, SA) equipped with reversed-phase analytical column C18 (Poroshell 120 EC-C18, Agilent Technologies). Linear gradient elution was carried out with a solution of 10 mM potassium dihydrogen phosphate. Among the studied analytes, allantoin showed the greatest concentration, averaging 3071.10 µg/mL, followed by creatinine and uric acid, averaging 505.91 and 225.05 µg/mL, respectively. The strongest Pearson's correlations were observed between allantoin and creatinine ($r = 0.76$; $p < 0.001$). Uric acid was moderate-to-low positively correlated with allantoin ($r = 0.31$; $p < 0.001$), and creatinine ($r = 0.28$; $p < 0.001$). Creatinine negatively correlated with milk yield ($r = -0.18$; $p < 0.001$) and weakly positively correlated with milk protein content ($r = 0.10$; $p < 0.05$).

The volatolomic signature differs between primiparous and multiparous cows across different matrices

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Non-invasive phenotyping of cattle is becoming of increasing importance to develop new tools to study and individually manage farm animals. We explored the effect of the dairy cows' parity on the volatolomics of several non-invasive matrices such as breath, ruminal liquid, urine, faeces and sweat. Ten primiparous (PRIM, 73 ± 11 days in milk, 22.3 ± 3.31 kg/d milk yield, 618 ± 41 kg BW, 1.5 ± 0.20 BCS) and 6 multiparous (MULT, 2nd lactation, 23.0 ± 1.59 kg/d milk yield, 76 ± 11 days in milk, 642 ± 49 kg BW, 1.4 ± 0.20 BCS) Holstein cows were fed ad libitum a high-fibre diet (53% FDN, 12% CP) during 5 weeks. Cows were sampled at the end of weeks 4 and 5, and the samples were stored until analysis (-80°C) except for breath, which was stored at room temperature and analysed within 24h after sampling. Samples were thawed (37°C, 10 min), heated (45°C, 30 min) and their headspace analysed through selected-ion flow-tube mass spectrometry (SIFT-MS) in full mode scan (positive and negative ions). The parity effect on the volatolome was explored through Partial-least square discriminant analysis considering weeks 4 and 5 independently. The breath volatolome discriminated the parity only in week 4 ($R_2Y=0.96$, $Q_2=0.83$) being the best discrimination model. The faeces did not yield any valid model. The rest of the matrices always yielded a valid model, with acceptable predictive ability observed for ruminal liquid on week 4 ($Q_2=0.59$), and urine ($Q_2 = 0.58$) and sweat ($Q_2 = 0.59$) on week 5. Our results suggest that metabolic and digestive differences between primiparous and multiparous lactating cows were caught by the volatolome of these matrices, and so future studies should consider the effect of the parity.

Individual response of dairy cattle to heat waves: phenotypic and genotypic characterization

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This study aimed at evaluating the impact of heat stress exposure on milk production patterns in terms of milk decline and recovery, and length of stress. Individual daily data were collected from 1176 cows in seven herds located in Arborea (OR, Italy). The farms were equipped with automatic milking systems and the data spanned from January 2022 to August 2023. In correspondence with heat waves occurrence, percentage of milk lost (MYL), milk recovered (MYr), and the stress duration length (SL) in respect to previous performances, were estimated using a linear regression model, with farm and parity as predictor variables. Cows were genotyped with a medium density SNP bead chip. Heritability (h^2) for the three traits was estimated. A GWAS was also performed. Statistically significant differences ($p < 0.001$) were observed in MYL, SL, and MYr metrics among the seven herds. The MYL ranged between $-29.95 \pm 1.57\%$ and $-6.96 \pm 1.77\%$, SL between 12.4 ± 0.39 and 14.0 ± 0.30 days, MYr between $+17.63 \pm 1.51\%$ and $+3.68 \pm 1.53\%$, respectively. Parity significantly affected SL ($p < 0.001$) and MYr ($p < 0.05$). First and third calving cows showed the highest SL (13.0 ± 0.27 days), whereas cows at fifth calving exhibited the lowest (12.7 ± 0.39 days), respectively. Moreover, fifth parity cows showed displayed the highest MYr of $+15.7 \pm 2.03\%$, whereas fourth parity exhibited the lowest MYr of $11.3 \pm 1.74\%$. Two SNP were associated with MYL (located on BTA14 and BTA19), one with MYr (CBTA23) and 29 with SL. The estimated h^2 values for MYL, SL, and MYr were 0.08 ± 0.05 , 0.07 ± 0.04 , and 0.04 ± 0.03 , respectively, with corresponding repeatability of 0.15, 0.12, and 0.11.

Session 13

Theatre 6

Supplementing Bovacillus to weaned beef steers during a 90-day grazing period

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This experiment compared performance, health, and physiological responses of beef steers receiving a *Bacillus subtilis* probiotic during a 90-day grazing period. Steers ($n = 240$) were obtained from an auction yard and transported to the experimental facility. Steer shrunk body weight (BW) was recorded at arrival (day -1). This value was averaged with BW recorded on day 0 to represent initial BW (237 ± 1 kg). On day 0, steers were ranked by initial BW and allocated to 1 of 24 pastures (4-hectare pastures; 10 steers/pasture). Pastures were randomly assigned to receive daily supplementation with dried distillers' grains at 1% of BW containing either: 1) *B. subtilis* ingredient (BOV; 2 g/steer daily of Bovacillus™; Chr Hansen A/S, Hørsholm, Denmark) or 2) no additive (CON). Steers received treatments from day 0 to 90 while housed in pastures containing stockpiled native grass forage. Steers were assessed for bovine respiratory disease (BRD) signs daily. Blood samples were collected, and BW recorded on days 0, 14, 28, 56, 90. Shrunk BW was recorded on day 91 for average daily gain calculation (ADG). Treatment x day interactions were detected ($P < 0.05$) for plasma concentrations of cortisol and parainfluenza type 3 (PI3) antibodies. Steers receiving BOV had greater cortisol on day 56 ($P < 0.01$) and also greater ($P < 0.01$) PI3 antibodies on day 90 compared to CON. No treatment differences were detected ($P = 0.97$) for BRD incidence, although steer mortality + removals for health complications were greater in CON compared to BOV (5.0 vs. 0.0%; $P = 0.01$). Body weight and ADG did not differ between treatments ($P \geq 0.24$). Total liveweight change per pasture was greater in BOV compared to CON (645 vs. 501 kg liveweight change/pen; $P = 0.04$). Despite lacking difference for incidences of BRD or physiological responses, an increased number of BOV steers completed the trial. Therefore, *B. subtilis* supplementation increased overall production liveweight in weaned beef steers during a 90-d grazing period.

Development of a proxy to estimate individual daily DMI of Belgian Blue bulls using fecal NIR spectra

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Optimizing feed efficiency of Belgian Blue (BB) bulls is one of the aims of the “Blanc Bleu Vert” project (SPW Research funding-Wagralim) to help the beef sector to reach European climate objectives. To reach this goal, the aim of this study is to develop an indirect and robust tool applicable in routine settings (proxy) to predict individual dry matter intake (DMI) using fecal near-infrared spectrometry (NIR). Spot samples of feces were collected from 288 BB fattening bulls (aged from 9 to 20 months) every 3 weeks and their feed intake was measured daily (7.9 ± 1.4 kg DMI/day) resulting in a total of 861 combinations of reference data. Using partial least squares (PLS) regression, different combinations were tested regarding the DMI values considered in parallel with each spectrum: daily or average DMI from 1 to 7 days before fecal sampling. Furthermore, weight, abdominal perimeter, and diet information were considered individually or combined as predictive values in addition to spectral information. At this stage, the reference dataset exhibits a lack of variability, with only one breed represented, animals at the same physiological stage, and all receiving the same dry diet. Consequently, the statistical results are unconvincing (SEC=1.1 kg DMI/day). In the coming months, new reference data will be collected regularly from 94 additional BB fattening bulls (culard or crossbred) fed dry or fresh diets (based on corn and/or grass silage). This expanded variability in the calibration set is expected to enhance the robustness of the prediction model.

Early post-mortem discrimination between grass- and grain-fed beef cattle using muscle lipid biomarkers

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The aim of this study was to create a muscle lipid-based biomarker panel to discriminate between grass-fed and grain-fed beef cattle early post-mortem. Carcasses from grass-fed ($n = 27$; from 30 to 35 mo age) and grain-fed ($n = 34$; from 20 to 24 mo age) Nellore (*Bos Indicus*) bulls were obtained at a commercial slaughterhouse. Longissimus thoracis muscle (11th rib level) samples were collected at 30 min post-mortem for further lipidome analysis using multiple reaction monitoring (MRM)-profiling. Principal component analysis (PCA) and clustered heatmaps were performed. Moreover, receiver operating characteristic (ROC) curve analysis was performed to create a predictive model for evaluating the performance of putative biomarkers. Out of the 1366 ion transitions (MRMs) scanned, 300 had intensities of at least 1.3-fold higher than the blank and were considered for statistical analysis. The PCA and clustered heatmap analyses revealed distinct clusters between the muscle lipidome from grass- and grain-fed cattle. Based on the ROC curve, a model was created to predict the animals' feeding regimen, which obtained a maximum predictive accuracy of 96.2% using only 15 putative biomarkers, misclassifying only 2 out of the 61 samples. Muscle lipidome from grass- and grain-fed beef cattle has a very discriminating profile, which allowed creating a highly predictive model to discriminate them during early post-mortem. Therefore, this chemical test can be used as a definitive tool to exclude any possibility of fraud/adulteration in relation to the animals' feeding regimen, increasing the reliability of the grass-fed beef certification.

Continuous monitoring of glycaemia to detect variations in intermediary metabolism during night and day cycles in growing pigs

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Monitoring time-course variations in glycemia is important to depict energy metabolism which allows the animal to cope with challenges. The aim of this study was to monitor continuously glycemia in growing pigs to depict inter-individual variations in intermediary metabolism along day and night cycles. For that, smart glucose sensors estimating interstitial glycemia at 5 minutes interval as a substitute for circulating glucose concentrations, were secured on the neck of female pigs (n = 7; Pietrain x (Large White x Landrace), 50 kg live weight) that were housed in individual cages during 5 days and fed twice a day. Two meal test procedures were applied during the trial, one with a high-starch diet and the other one with a high-fat diet. Spearman correlations were used to explain glycemia of the day by glycemia of the night. On average, there were 3.2 + 1.8 (mean + SD) hyperglycemic peaks and 3.3 + 0.8 hypoglycemic peaks during the day, and 1.6 + 0.5 hyperglycemic peaks and 2.0 + 0.9 hypoglycemic peaks during the night when pigs had no access to feed. When pigs were fed the high-starch diet, the number of nocturnal hyperglycemic peaks was negatively correlated with the number of diurnal hypoglycemic peaks ($r = -0.93$, $p < 0.01$). Duration of hyperglycemic peaks in the night tended to be positively correlated with duration of hyperglycemic peaks during the day ($r = 0.75$; $p = 0.08$). Peak glucose concentration in the night was positively correlated with the duration of diurnal hypoglycemic excursions ($r = 0.82$, $p = 0.03$). Peak glucose concentrations during the night were lower when pigs were fed a high fat diet than those observed when pigs were fed the high starch diet (135 mg/dL vs 155 mg/dL on average, $p < 0.05$). Overall, this study opens new perspectives to synchronize feeding frequency with day/night cycles to optimize energy use in precision feeding. The PigWeb project has received funding from the European Union's Horizon 2020 under the Grant Agreement N° 101004770.

Session 13

Theatre 10

Faecal microbiota and stillbirth rate in sows

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Stillborn piglets are of increasing concern for the pork industry, with losses ranging from 5-20% (Langendijk & Plush, 2019). The gut microbiota has been shown to contribute to sow productivity, including stillbirth (Wang et al. 2019). Microbiota manipulation via in-feed probiotics may reduce stillbirths, however further research is needed to identify which bacteria to target. The aim of this study was to identify potential microbial drivers for stillbirth in sows. Faecal samples from 87 sows were collected on entry into conventional farrowing crates, 4.1 ± 0.1 days prior to farrowing. Parity was recorded, and sows were characterised as having given birth to none, one, or more stillborns. DNA from the faecal samples was subjected to 16S rRNA gene amplicon analysis. Sequence data was processed to ASVs in QIIME2 and analysed and visualised with MicrobiomeAnalyst (<https://www.microbiomeanalyst.ca/>). Sows with stillborn piglets had significantly lower microbial diversity than those sows with no stillborns ($P < 0.05$). Tepidbacter genera were more abundant in sows with stillborn piglets, while Megasphaera and Dialister were more abundant in sows that had no stillborns. Megasphaera and Dialister were in highest abundance at parity 3 and declined slightly by parity 6, with larger variability between sows. These bacteria are candidates for the development of an in feed probiotic to enable a targeted approach for reducing stillbirth. References: Langendijk & Plush. 2019. *Animals* 9:885./ Wang et al. 2019. *Theriogenology* 136:131-137.

Prediction of the daily nutrient requirements of gestating sows based on behavioural sensor data and machine-learning algorithms

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Precision feeding is a strategy for supplying feed as close as possible to each animal's nutrient requirements. Usually, the nutrient requirements of gestating sows are provided by a model using input data such as sow characteristics as well as an estimation of future farrowing performances. New sensors and automations have been developed on pig farms over the last decade, generating large amounts of data. This study proposes to predict daily nutrient requirements, using data measured by sensors recorded on 73 gestating sows. Considering various digital farm configurations (for example electronic feeders and drinker stations), we explore and evaluate the performance of nine machine-learning algorithms to predict the daily metabolizable energy and standardized ileal digestible lysine requirements for each sow. Their prediction results were compared to those predicted by the InraPorc model, a mechanistic model designed for precision feeding of gestating sows. The higher correlation coefficient values for lysine (0.99) and for energy (0.95) were obtained for scenarios involving automatic feeder system alone or combined with another sensor. For the scenarios using data from the automatic feeder only, the root mean square error was lower with gradient tree boosting (0.91 MJ/d for energy and 0.08 g/d for lysine) compared to those obtained using linear regression (2.75 MJ/d and 1.07 g/d). The results of this study show that the daily nutrient requirements of gestating sows can be accurately predicted using data provided by sensors and machine-learning algorithms. This paves the way for simpler solutions in precision feeding.

Comparison of stress markers for measuring heat stress of meat type ducks

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Heat stress is a major concern in the poultry industry because it is closely related to productivity and animal welfare. Therefore, research on measuring heat stress and developing appropriate indicators must be conducted. This study aims to perform a comparative analysis of the effect of rearing seasons on stress markers [corticosterone (CORT), Heat shock protein 70 (Hsp70), H/L ratio] in meat-type ducks. The treatments were categorized into summer and fall. A total of 100 ducks were used for this study. Stress markers were analyzed in the blood and feather. All data were statistically analyzed using the independent t-test procedure of SAS. Blood Hsp70 levels were higher in fall (1.56 ± 0.63) than in summer (1.36 ± 0.44) ($p > 0.05$), but it was no significant difference. And blood CORT levels were significant higher in fall (1634.07 ± 967.96) than in summer (946.53 ± 632.00) ($p < 0.05$). The H/L ratio was higher in summer (1.31 ± 0.69) than in fall (1.06 ± 0.86) ($p < 0.05$). Feather CORT levels tended to be higher in summer (6.73 ± 2.47) than in fall (6.60 ± 2.37) ($p > 0.05$), but it was no significant difference. And feather Hsp70 level was significantly higher in summer (0.46 ± 0.16) than in fall (0.20 ± 0.13) ($p < 0.0001$). Blood CORT is highly affected by capture and sampling time, it can be difficult to interpret the CORT measurement under some circumstances. Feather Hsp70 is a long-term stress marker and is more sensitive to heat stress than blood. The H/L ratio is influenced by injuries or diseases. Therefore, it should be determined multiple times in other areas of the body to enhance accuracy. Among other stress markers used, feather Hsp70 was assumed to be the most sensitive indicator of heat stress. The data can be used as basic for developing high-temperature stress indicators and complementary research.

A meta-analysis assessed factors affecting milk components and intake in response to metabolizable methionine fortification using INRA 2007

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We aim to establish response equations on milk fat contents (MFC), yield (MFY), and milk yield (MY) to digestible methionine (MetDi) supplementation as Smartamine M or MetaSmart and to identify secondary influencing factors. Meta-analysis was applied on 25 peer reviewed papers. Dairy cows consumed 20.3 ± 4.1 kgDM, 172 ± 18.2 g CP/kgDM, 1643 ± 70 MJ NEL/kg DM and $2.35 \pm 0.34\%$ MetDi. Models showed a 0.7 %MetDI increase resulted in a 0.12% MFC ($R^2=0.91$, RMSE=0.10) and 84 g/d MFY ($R^2=0.97$, RMSE= 52.1) increase. Within-study slopes of models were negatively impacted by fermented OM (FOM; $P<0.01$), starch degradability (dSt) and IleDI ($P<0.1$) and positively by lactation stage, DMI and fat content ($P<0.05$) for MFY, and were positively impacted by DMI, and NEL ($P<0.1$) for MFC. For a same MetDI, MFC increased in early lactation with DMI, starch, and decreased with NDF, dSt, FOM, ThrDI, ValDI, IleDI and GlyDI contents ($P<0.01$). Models showed also an increase of 0.48 kg milk/d ($R^2=0.98$, RMSE=0.99) and 0.49 kg DM/d ($R^2=0.98$, RMSE=0.57) by increasing 0.7%MetDI. Within-study slopes were positively impacted by lactation stage ($P<0.05$) and negatively by FOM ($P<0.01$) for MY model and positively by concentrate and ProDI ($P<0.1$) for DMI model. For a same MetDI, MY increased with DMI, fat content and decreased with NDF, dSt, FOM, ThrDI, ValDI, IleDI, PheDI and GlyDI contents ($P<0.01$), and DMI is increased by NEL and decreased by dSt and FOM ($P<0.01$). Predicting dairy cows' responses to MetDi and understanding influencing factors is feasible, enhancing comprehension and aiding dietary formulation improvements.

Session 14

Theatre 1

Moo-ving forward: AI's role in transforming dairy calf care

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Diarrhea and Bovine Respiratory Disease complex (BRD) are the leading causes of morbidity and mortality for dairy calves so timely disease detection tools are needed. This presentation explores precision livestock farming technologies (PLF) and artificial intelligence (AI) to detect clinical disease status in dairy cattle. Feeding behavior data recorded from robotic milk feeders/concentrate feeders, activity behavior from pedometers, and temperature monitoring technologies will be reviewed as strategies to identify sick calves. Furthermore, this presentation explores different PLF alerts for sick calves, including averages of sick animals vs. individual calves as disease detection strategies. Last, this presentation covers machine learning, AI, and PLF as a disease detection strategy for calves. Briefly, Cantor and colleagues have observed that PLF technologies can capture a pre-clinical calf for diarrhea and BRD, as well as if a calf will recover or not from antibiotic treatment for BRD. Furthermore, feeding non-antimicrobial treatments (e.g., colostrum replacer 0.13 kg/d for 3 days by bottle) to older calves that deviated from their baseline feeding behavior measured by a robotic milk feeder reduced the hazard of BRD status. Specifically, control calves had a 2.4 greater hazard of BRD within the next 15 days compared to colostrum calves (95% CI: 1.3-4.3). They have also observed that machine learning techniques and AI are powerful classifiers: a KNN algorithm had 96% accuracy for labeling BRD calves using pedometer and robotic milk feeder data at -5 days before clinical diagnosis. Since most producers cannot afford multiple PLF systems, an economic AI scenario was also considered. Even with budget sensitive algorithms, using one PLF system improved BRD detection accuracy and labeled BRD sooner compared to manual labor only. However, not all PLF technologies are equal. Cantor emphasizes that PLF and AI can improve calf care, but validation studies and alerts must be disease specific. Future research should develop a PLF BRD alert, and test additional non-antimicrobial intervention strategies for calves.

New directions on precision management for dairy cows

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Individualized feeding strategies have been explored to improve dairy cows' energy status. However, rules for precise and short-term adjustments at the individual scale still need to be defined. In a recent study, this individual concentrate adjustment was done weekly based on body weight gain without affecting the performance of the animals during the first fourth months of lactation. However, cows' metabolic status affected their performance and should be integrated in the decision rules of concentrate distribution. It can be predicted accurately during the first days of lactation based on three blood metabolites (glucose, BHB, NEFA). Moreover, studies suggested that delaying insemination after peak lactation could enhance pregnancy rates and reduce the number of inseminations per gestation. Health risks associated with peak lactation as well as management costs could be reduced as the number of calvings per cow per year decreases. Simulations suggested that especially primiparous could benefit from this strategy to optimize performance and that late insemination could improve cow longevity. To predict the consequences of combined reproductive and feeding strategies, prediction models are essential. The InSiliCow model can serve as digital twin to simulate dairy cow's lifetime performance. It has been evaluated on different breeds, parities, lactation durations and feeding strategies. An on-going project is aiming at improving it and using it to identify optimal feeding and reproductive strategies to enhance cow performance and welfare, considering available technologies. This involves determining the energy content, level, and frequency of ration adjustments, as well as variables for adjusting rations and insemination timing.

Session 14

Theatre 3

Improved monitoring with historical data, expert knowledge and context measures integration applied to dairy cow heat stress

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Optimizing animal health and welfare is crucial to improve animal production sustainability. Digitalization offers elegant opportunities to improve livestock monitoring with limited (human) resources. The complexity of the animals and their production context are the main bottlenecks to achieving valuable results. We propose maximizing data use and optimizing decision making in livestock monitoring by integrating different data sources and modelling techniques. Specifically, our project aims to develop solutions for animal monitoring by explicitly considering (1) historical data, (2) expert knowledge, and (3) context and control measures. We apply these to dairy cow heat stress, using multiple years of activity and milk yield data from 11 farms (BE, NL, IT) with a Lely milking robot and the nearby weather. Historical data are considered by assuming (animal-individual) prior time series trajectories and distributions. Expert knowledge is integrated via smart feature design, e.g. including time lags and recovery capacity from warm temperatures. Context and control measures are entered in the modelling explicitly, thereby distinguishing between herd and individual effects. With this methodology, we found that up to 61% of the daily individual variability in milk yield and activity can be explained by outdoor climatic conditions, and that the inclusion of time-lagged and recovery features is essential to reveal individual-cow sensitivity to heat stress. This method can be used on farm to predict heat stress sensitivity or design precision phenotypes.

Integrating IMU wearable sensor data for enhanced PLF in dairy calves

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The growth of wearable sensors in Precision Livestock Farming (PLF), highlights the need to understand the data output fundamentals and values they produce once integrated with other data. Using raw output recording at 10Hz produces 864,000 records per day, thereby providing the ability to gain deeper insight and analysis. Working with four 20-hour recordings from neck-based Inertial Measurement Units (IMUs) on two, four-week-old calves, the primary objective was to assess state-based annotation (lying vs standing) with raw data outputs and identify its value for Machine Learning (ML) model development. The IMU raw data was annotated marking start and end times, with synchronized video footage and appended to the sensor data. After annotating states of IMU data, rolling 1, 5, and 10-second cumulative movement summaries were appended to improve ML model development. The model trained on three datasets using Python's scikit-learn library's Random Forest Classifier and tested on the fourth dataset to predict states. Preliminary results average 93.1% accuracy (correctly predicted states over total observations within the test dataset) with the limited test data. Artificially slowing the recording frequency to 1Hz and 0.1Hz lowered accuracy to 87.2% and 86.0%, respectively. Reducing the data to the 3 acceleration axes output, the accuracies were 80.9% (10Hz) 80.1% (1Hz) and 79.5% (0.1Hz). Growing the library of annotated data will improve model development as future research with other ML approaches and will benefit from larger raw output datasets. This method displays the value of IMU wearable sensor raw data outputs integrated with video-based annotation for use in PLF behavioral research.

Session 14

Theatre 5

Designing a local-level data space platform as a strategic approach to reduce human-wildlife conflicts in rural Italy

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Human-wildlife interaction is a key aspect of landscape management, posing significant challenges encompassing issues such as biosecurity, hunting regulations, and farm damage. Currently, these challenges are fragmented across different public administration offices, making coordination extremely difficult. This study utilizes human-centric feedback to design and deploy an AI-driven platform to bridge the information gap. Based on stakeholder analysis, a data space platform was designed aiming to enhance real-time monitoring of field situations and provide early feedback on implemented policies. The stakeholder analysis involved semi-structured interviews (n = 12), which were transcribed, evaluated using direct semantic methods and scored for keywords and prioritization of challenges. Their analysis revealed a highly fragmented nature of decision-making within public administration, with different offices holding divergent viewpoints and interests. Therefore, a data space design was chosen, which relies on decentralized data gathering to ensure data sovereignty for individual offices. Additionally, gradual onboarding of data sources was developed to simplify early-stage validation. In conclusion, the design process for an AI-driven data space for relevant administration problems involved both human-centric and technical assessments of suitable AI methods. Such solutions have the potential to significantly streamline the decision-making process in the management of complex human-wildlife interactions at the landscape level.

Quantifying beef cattle grazing behaviour using Sentinel-2 data in South Africa's Strandveld region

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Remote sensing has become an increasingly popular tool to monitor grazing behaviour and aid in rangeland management and conservation. It is well-known that vegetation quality and composition influence livestock grazing behaviour. This study evaluates various vegetation indices, including the Near Difference Vegetation Index (NDVI), for predicting beef cattle grazing behaviour in South Africa's Strandveld region. Six Bonsmara (*Bos taurus indicus*) and six Nguni (*Bos taurus africanus*) cows were each fitted with a LoRa (GPS/UHF) collar and placed on the natural vegetation of Nortier Research Farm, Western Cape, South Africa. GPS coordinates of cattle locations were recorded for 12 months. Statistically significant spatial clustering of GPS data was identified through Optimized Hot Spot Analysis. Hot- and cold spots were identified, indicating that cattle preferred certain paddock areas. In contrast, others were visited significantly less frequently. Hotspots were statistically compared to values extracted from Sentinel-2 imagery using various vegetation indices. It was observed that NDVI data on vegetation quality could not fully explain cattle grazing behaviour and that other Sentinel 2-derived indices needed to be considered when attempting to explain and predict grazing behaviour. Remote sensing technologies are valuable tools for rangeland management; however, a one-size-fits-all approach cannot be followed, and various indices must be evaluated for different vegetation types.

LiDAR technology as a contactless tool for quantitative analysis of body condition score in beef cattle

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Body condition score (BCS) is a crucial aspect of livestock management being a key indicator of productivity, health and welfare of animals. Modern contactless techniques are constantly being implemented for an increasingly effective BCS assessment. Light Detection and Ranging (LiDAR) uses laser scanning and point-to-point matching to construct 3D shapes that accurately represent the animal's body pattern. Therefore, LiDAR represents an opportunity for a cost-effective and accurate shape computation. The current research aims to assess LiDAR technology's reliability in beef cattle. LiDAR tool by both mobile phone and unmanned aerial vehicle will be used to get 3D point cloud of beef cattle, deriving the critical parameters to estimate BCS. Simultaneously, the BCS will be evaluated by a breed classifier following the standard method. The outcomes from both methods will be compared to assess the performances and accuracy of the contactless LiDAR approach. 3D data processing for animals will be improved and specific algorithms will be developed to facilitate the automatic discrimination between the BCS classes and also exploring the possibility of computing animals' weight in the future. A more reliable, accurate, and timely predictions of cows' BCS can improve some critical aspects of beef cattle farming, such as farm management, genetic evaluations, and animal welfare, bringing significant advantages for farmers.

Early lactation body condition and milk production affects fertility performance in dairy cows

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Improving dairy cow fertility is difficult, as it is a complex trait that depends on the animal's physiological status, health, and production performance. How a cow navigates through her transition period at the start of lactation, has been shown to affect fertility performance later in the lactation. With sensors that can reveal physiological status over time at a high frequency, we can study the complex relationships between early lactation health and production, and fertility performance. In this study, we use an extensive dataset from 17 commercial dairy farms that are equipped with an automatic milking system, a milk progesterone monitoring sensor and, for 5 out of 17 farms, a body condition score (BCS) camera. Meaningful features were derived from the early lactation BCS and milk production time series. These features describe for example changes in BCS during the dry period and early lactation, total milk yield and milk yield variation. Random forest regression models were used to link the BCS and milk production features to fertility performance traits derived from the milk progesterone time series. Based on the fertility trait and features included in the model, the r^2 ranged between 0.05 and 0.15. We found that a prolonged postpartum anestrus period was associated with a stronger decrease in BCS during early lactation. A prolonged period from calving to successful insemination was associated with high cumulative milk yield and high milk yield fluctuations during early lactation. These insights can be used to better understand the factors that jeopardize dairy cow fertility.

Session 14

Theatre 9

Digital and precision livestock farming (PLF) technologies: gaps between sheep and goat farmers' needs and existing solutions

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Sm@RT is an EU Horizon 2020 funded project, involving Estonia, France, Hungary, Ireland, Israel, Italy, Norway and UK, to encourage uptake of digital and PLF technologies by sheep and goat farmers. In a series of national and international workshops in the 8 countries, 166 different needs and challenges regarding technology use were identified by sheep and goat farmers. Sixty potential solutions were collated by the project partners, with some solutions addressing more than one need and production type (meat sheep, dairy sheep and dairy goats). In total, 18 needs were not addressed as follows: 3 for reproduction, 4 for health/welfare, 3 for herd monitoring, 2 for feeding/grazing, 4 for milking, and 2 for fattening. During a final international workshop, additional solutions were identified by over 80 stakeholders to address those gaps although not all were available on the market or directly adapted. Only two gaps relating to milking were not addressed. Between existing technologies and upcoming prototypes, most sheep and goats farmers' needs for technology use on farms can be answered. However, barriers to uptake and cost-benefit analysis, amongst others, should also be considered.

Using accelerometers for goat activity monitoring

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Monitoring animal activity is crucial for welfare, health, and has potential future economic impact. Although the cost of accelerometers remains high, they prove particularly useful when the objective is to monitor the activity of animals raised in groups in a research setting. The aim of our study was to develop an algorithm to predict animals' activity based on Ax3 accelerometers from Axivity, known for their impressive battery life and lack of time synchronization issues. We established an experimental pasture covering approximately 150m², where 4 to 5 adult goats grazed freely for 1 to 2 days. The animals were equipped with accelerometers attached to their horns. CCTV cameras were used to film the animals during the experiment, allowing for manual labeling of their behavior afterward. A total of 35 behaviors were considered, including generic actions such as ruminating or grazing, as well as more specific behaviors like grooming or fighting. The animals' activities were manually labeled from 72.44h of video footage using Boris. For the behavior, we try a deep-learning approach based on LSTM. We proposed a waterfall model, i.e. an iterative LSTM model, with different sequence lengths, adapted to the each tested behavior. For prediction, we only considered 5 behaviors: displacement, grazing, ruminating, other and resting. The recall was 52.9%, 99.4%, 92.9%, 79.2%, and 95.5%, while the precision was 70.8%, 99.1%, 89.8%, 86.7% and 96.2%. The effectiveness of the method was lower for shorter behaviors, partly due to the challenge of perfectly synchronizing video and accelerometer data.

IntelliRAS – some results from a project on precision fish farming in recirculation aquaculture systems

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IntelliRAS, “Intelligent farming and health control in land-based recirculating aquaculture systems” (2021-2024), is a Nordic project including three research institutes and a Danish company producing and selling probes measuring e.g., dissolved oxygen (DO; amount of oxygen accessible to the fish) and CO₂ concentration in aquaculture systems. The overall aim is to enable farmers producing salmonids in recirculation aquaculture systems (RAS) to move from experience-based to knowledge-based decision making; precision fish farming (PFF). PFF was introduced in 2018 to facilitate a more sustainable production regarding production margins, environmental impact, health, and welfare in intensive aquaculture. Here we focus on monitoring and predicting mortality and consider tools for management that may enable earlier and less radical preventive changes in feeding. The methods considered are quality control charts of daily mortality counts, forecasting of CO₂ and DO based on sensor data time series, and use of generalised dynamic linear models. The main data are from a Danish land-based freshwater trout farm with 12 tanks, 2 with additional sensors measuring DO, CO₂ and temperature in the water. Moreover, we have production data from ~15 Danish RAS trout farms. Preliminary analyses showed CO₂ to be in moderate positive correlation with feed fed but uncorrelated with mortality, which had positive moderate correlation with temperature.

Optimizing Disease Risk Classification: A Data-Integrative Approach

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In this study we apply a data integrative modelling approach and machine learning (ML) techniques to advance the classification of dairy cows at health risk. Central to our approach is the optimization of the balance between static and dynamic information and an evaluation of the interplay between model inputs and disease classification performance. By synthesizing insights from previous research on farm and livestock risk models, we minimize reliance on static traits by highlighting the most important traits, improving the application of an integrative approach to disease risk modeling. Analyzing data from 342 farms and 24,663 dairy cows, our study integrates over 350 static features, including farm management strategies and genetic information, alongside dynamic features such as DHI milking records and AMS data, and health diagnostic outcomes. A variety of machine learning techniques, such as XGBoost and RF, are applied to identify the best classification model. Preliminary findings demonstrate the efficacy of the XGBoost model in predicting lameness, achieving a high degree of sensitivity (0.81) and specificity (0.94), culminating in an F1 score of 0.78. This underscores the potential of employing advanced ML techniques in the precise identification and management of health risks in dairy cows. The study represents a further step towards the development of a data-integrative approach to disease risk modeling that supports farmers in their decision-making process.

Assessing the future uptake of digital tools and technologies by sheep and goat farmers in the meat and dairy industries

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Sm@RT, an EU Horizon 2020 funded project, aims to improve the understanding, awareness and uptake of digital tools and technologies available to small ruminant farmers. To assess factors influencing the rate and peak level of uptake, stakeholder groups in the 8 Sm@RT countries (Estonia, France, Hungary, Ireland, Israel, Italy, Norway, UK) used the Adoption and Diffusions Outcome Prediction Tool (ADOPT) (<https://adopt.csiro.au/home.aspx>). Questions were in 4 categories; characteristics of the tool/technology; characteristics of the farming population; advantage of using the tool/technology; and learnability. To date, sessions have been completed on 20 different tools/technologies presented by the project as possible solutions to needs identified previously. For example, predicted rate and peak level of uptake for an EID stick reader (across 4 different countries) ranged from 9-24 years and 72-97% of the population. Overall results indicate that answers given relating to what proportion of the population will need new skills/knowledge, and the proportion of farms that could benefit from the tool/technology, are influential in terms of rate and peak level of uptake respectively.

Web platform for technical and economic comparisons of environmentally beneficial practices and investments in pig, poultry, and cattle farming

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In the context of reducing the environmental impacts of livestock farming, IFIP-Institut du porc, supported by six partners, has developed a web platform for technical and economic comparisons of environmentally beneficial practices and investments in pig, poultry, and cattle farming. The main objective of this tool is to guide farmers towards the most competitive environmental practices, e.g. those with the best efficiency/cost ratio. A second objective is to create national benchmarks for updated costs. The aspects considered include storage, treatment, and spreading of livestock effluents (slurry, manure, digestate); reduction of ammonia emission; techniques for reducing energy consumption and producing renewable energy. In practice, farmers create an account on this platform and fill in a technical description form of the service or investment made on one of the mentioned items and the cost invoiced to them. Participation in the program allows each farmer to compare their costs to a similar group and identify the reasons of the observed differences. Filters, consisting of the main technical descriptions (capacity of the unit, installed power, number of animals or mass of manure involved, etc.), allow the farmer to specify or broaden the reference population to which they compare themselves. To initiate the system, the technical and economic data of more than 40 farmers on one or more aspects have been integrated into the tool.

CowBase: a Python package for streamlining data management in dairy farming

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With the increased implementation of automatic milking systems (AMS) and the trend toward larger farms, translating data into useful information has become crucial in the dairy sector. Although there is increasing research on understanding and analyzing dairy data, the absence of standardized practices among research groups and the time-consuming nature of building proper data structures present significant challenges. In response, we present CowBase, a Python package designed to streamline data management from AMS and weather stations. The first two of five modules extract data from an AMS farm backup or directly from an on-farm database, with current implementation for both DeLaval and Lely systems. The third module allows for data selection and contains functions for robust data processing, optimizing data tables for research purposes, and correcting for data discrepancies. Finally, the fourth module generates and writes to a new data structure, supporting various database systems such as PostgreSQL, MySQL, and SQLite. In the last package, a database connection class for Python is given. Testing and validation of CowBase were done on data collected in projects within the Livestock Technology group at KU Leuven, resulting in a database of 60 dairy farms, with data from back to 2005, facilitating over 10 PhD projects and collaborations. By providing a comprehensive solution for data management in dairy farming, CowBase aims to improve efficiency, facilitate collaboration, and advance research in the field.

Improving conception rates for farms with a milk progesterone sensor

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The use of milk progesterone (P4) has become the gold standard in dairy cow fertility management as it allows to accurately identify the optimal insemination window based on the detection of luteolysis preceding ovulation. On-farm P4 devices (OPD) measure and monitor the milk P4 of individual cows, thereby generating luteolysis alerts (LA) and estimating the optimal insemination window. Currently, a multi-process Kalman filter is used by the OPD to process the raw P4 data and reduce false LA. This filter introduces a time lag on the LA which depends on measurement frequency and the rate of change in P4 during luteolysis. The variation in this time lag is currently not considered in the optimal insemination window. In this study, we use an extensive dataset from 17 commercial dairy farms equipped with OPD. Conception rates for inseminations that are performed earlier, timely or later respective to the advised insemination were evaluated in function of the associated time lags. Results indicate that, when the time lag was longer than 24 h, conception rates can improve with 13% when inseminating earlier than advised. Likewise, when the time lag was shorter than 8 h, the conception rate increased 17.5% if the farmer inseminated later than advised. Our results suggest that farmers using OPD could increase their conception success by compensating a longer time lag on the LA with a shorter interval from alert to insemination. This will improve the fertility performance on those farms, which can positively impact the farm's sustainability.

Session 15

Theatre 1

Genotyping complex structural variants using a Chicken Pangenome reference

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Genomic structural variants (SV) comprise a sizeable proportion of the variable genome, but have remained difficult to characterize with short-read DNA sequencing technologies. It is necessary to still estimate the prevalence and impacts of these types of variants, so improvements in the efficacy of sequencing or the analysis of sequencing reads are required to assess the diversity of SVs in each species. Using a recently published Chicken Pangenome resource, we present the use of Pangenome graphs as a medium to efficiently identify SVs using short-read sequencing data. The Chicken pangenome graph was derived from 30 long-read chicken genome assemblies, with one previously published assembly (Huxu) of near telomere-to-telomere quality. The input assemblies provided excellent representation of the structural diversity of the Chicken genome when compared to previous studies, as a similar length of deleted sequence (19.2 Mbp previous; 19.7 Mbp Pangenome) and 3-fold higher length of inserted sequence (6.74 Mbp; 18.5 Mbp) were identified in the graph when compared to the bGalGal1b linear reference assembly. Having established that the graph was structurally representative, it was used as a reference to genotype complex structural variants, such as the K locus, in 100 sequenced chicken individuals. K locus genotypes were consistent with the phenotypes of the sequenced birds; however, the use of the graph revealed that 20% of birds with the late-feathering allele do not have a characteristic ev21 insertion in the locus, suggesting further structural plasticity at this locus. The use of a high resolution pangenome graph promises to improve the accuracy and throughput of SV detection in non-model organisms by resolving complex features that are recalcitrant to easy interpretation. If properly representative of the structural diversity of a species, a pangenome graph could allow for routine genotyping of SVs from cost-efficient short-read DNA sequencing.

Construction of a cattle pangenome for 14 French dairy and beef breeds provides new insights into their genetic diversity

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The current cattle genome reference assembly, based on a single Hereford cow fails to capture the whole spectrum of genetic variations within the species. Structural variations (SVs), defined as genomic variations longer than 50 nucleotides, can have a potential impact on both complex and Mendelian phenotypic variations. However they are difficult to detect using only standard approaches of either short or long-read sequence mapping to the current bovine genome assembly. Thanks to the recent advances in long-read sequencing technologies coupled with the development of appropriate bioinformatics tools, it's now possible to construct de novo genome assemblies for a large number of animals across various cattle breeds. It also offers the opportunity to study a broader range of both small and more complex genome-wide variations. Using these technologies, we have produced a comprehensive cattle pangenome incorporating genetic diversity from 64 high-quality de novo assemblies representing 14 French bovine dairy and beef breeds. We applied a combination of complementary approaches to characterize a wide spectrum of SVs and we report the identification of several megabases of novel genome sequences that are absent in the current cattle genome reference assembly. Further work is currently in progress to investigate the gene content of these non-reference sequences. This work was conducted in the SeqOccIn project, funded by the Occitanie region, FEDER, and Apis-Gen. Valentin Sorin's PhD is supported by INRAE.

Construction and characterization of a comprehensive ovine pangenome from 11 breeds provides new insights into their genetic diversity

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The current method of constructing a genome reference assembly through sequencing the entire genome of just one or a very small number of individuals falls short in capturing the complete spectrum of genetic diversity within most species, including ovine. In addition, the exploration of genomic structural variations (> 50 nucleotides) is constrained when relying solely on short-read sequences aligned to these genome reference sequences. Nevertheless, due to the rapid advancements in sequencing technologies and bioinformatics tools, it is now possible to generate long-read sequences and create de novo genome assemblies for numerous animals, thereby enabling the commencement of pangenomic studies within these species. In this study, we integrate long and short read sequences to build de novo assemblies and haplotype-resolved assemblies for 11 distinct ovine breeds. Employing various methodologies, we established a pangenome and subsequently investigated a wide range of structural variations within the species. We identified several additional megabases of genome sequences absent in the current reference genome assembly and we are currently investigating gene content of these non-reference sequences. This research was conducted as part of the SeqOccIn project, funded by the Occitanie region, FEDER, and Apis-Gen. Valentin Sorin's PhD is supported by INRAE.

Translocations associated with gonadal hypoplasia and colour-sidedness are common in northern Swedish cattle breeds

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The problem of gonadal hypoplasia, which can cause fertility problems, has been known in Swedish Mountain cattle (Fjällko) for almost 100 years. In the mid 20th century it was discovered that the inheritance was recessive with reduced penetrance and that the hypoplasia was associated with having a lot of white colour. Despite breeding efforts in Sweden during many years to reduce the prevalence of gonadal hypoplasia, the problem still occurs. Five Swedish cattle breeds, including Fjällko, show the phenotype of colour-sidedness which is caused by either a translocation and duplication from chromosome 6 to chromosome 29 (known as Cs29), or an additional translocation allele where part of Cs29 has been translocated back to chromosome 6 (Cs6). Besides the colour-sidedness, Cs29 is associated with gonadal hypoplasia. In this study, we used WGS data from 30 cattle of Swedish local breeds, as well as multiplex breakpoint PCR genotyping of additional cattle from different time periods to investigate Cs29 and Cs6. Analyses of the depth of coverage allowed us to detect the elevated depth of coverage created by Cs29 and Cs6. Within-sample allele frequencies, based on the depth of reads supporting each SNP allele located in the regions affected by the translocations, also support the inferred genotypes. We detected the Cs29 allele in all five Swedish breeds where colour-sidedness occurs. The allele frequencies of the Cs29 translocation ranged between 0.22 up to 0.57. We also detected Cs6 in four breeds. The genotyping of new samples showed that both translocations still occur in rather high frequencies.

Uncovering functional INDELS leading to alternative splicing in grazing sheep with different immune profiles exposed to gastrointestinal nematode infection

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This study aimed to identify insertion and deletions (INDELS) responsible for alternative splicing in Rideau x Dorset crossbred sheep with different immune profiles under natural exposure to gastrointestinal nematodes (GINs) using RNA-Sequencing. High (H) and medium (M) acute stress responders with the highest and lowest worm burden, after natural exposure to GIN, were selected for liver RNA extraction (H; n=5 and M; n=6). In addition, GIN-unexposed lambs (U; n=4) were used as immunological controls. Sequence reads were aligned to the annotated Oar_rambouillet_v.2.0 ovine reference genome. Quality control and variant discovery analysis were performed using CLC Bio Genomics workbench. Functional consequence analysis to identify INDELS leading to amino acid changes and splice site events (SSEs) were performed. Genes were recovered from Ensembl BioMart using the list of INDELS. Unique INDELS with SSEs for H, M, and U groups (162, 244, and 161, respectively) and shared (372) among groups were identified. Among them, 4 INDELS with SSE uniquely identified in the M group were located within the NLRC5, SIGLEC1, CFD, and STAT6 genes related to the immune system. Significant enriched metabolic pathways (FDR<0.05) were identified using the list of INDELS with SSEs for each group using Reactome. The cross-presentation of soluble exogenous antigens (endosomes) and NIK noncanonical NF-κB signalling pathways were identified in the M group. These results can help to better understand the mechanisms responsible for sheep's immune response to GIN.

SNP-based identification of Robertsonian translocation 1;29 in 32 Italian cattle breeds

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Robertsonian translocation 1;29 (rob(1;29)) is a chromosomal anomaly impacting cattle fertility. Screening programs have reduced its prevalence over time, but rely on cytogenetic analyses, expensive and requiring fresh blood. This work uses routine SNP data to explore the genomic region involved in rob(1;29) and identify carriers within Italian cattle breeds. The study encompassed SNP data for about 1000 individuals from 32 Italian breeds, 172 of which also karyologically tested. Supplementary analyses were conducted on SNP data routinely collected by nine breed associations for 1700 animals. FST analysis identified a region within the initial 6 Mb of BTA29, which includes the centromeric region implicated in the translocation. Principal component analysis and haplotype examination of this region proved to effectively detect heterozygous and homozygous carriers of rob(1;29). The study demonstrates SNP data's efficacy as a swift, precise, and cost-efficient tool for broad rob(1;29) screening, advocating for specific SNPs' inclusion in common cattle genotyping panels. The presence of a common haplotype among all identified rob(1;29) carriers supports a shared ancestry theory. Lastly, our analyses unveil the presence of carriers in previously untested breeds, with elevated prevalence -validated by karyological analysis- and the presence of homozygous individuals in the Cinisara, a breed of Podolian origin, which could serve as a model for investigating this translocation. Acknowledgements: The Authors are grateful to the BOVITA project and ANABIC, ANABORAPI, and ANAPRI associations for sharing their data.

Determining differential splice junction usage in the suckling lamb adipose transcriptome by comparing perirenal and tail fat

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Our study exploits transcriptomic data from perirenal and tail fat depots in suckling lambs (n = 8) to characterize alternative splicing (AS) events and differential isoform expression (DEI) in adipose tissue. Understanding isoforms' impact on adipose tissue development and fat deposition could offer valuable insights into metabolic regulation of the different fat depots analysed. We identified 4,446 differentially expressed genes (DEGs), with 2,156 overexpressed in perirenal fat and 2,290 in tail fat. Differential exon usage analysis unveiled 428 DEIs affecting 386 annotated genes, with exon skipping (~ 40%) and alternative transcription start sites (~ 35%) being predominant AS events. Of the DEIs within DEGs, structural changes were diverse, including alterations in intron structures, exon numbers, and genomic positions of open reading frames. Functional enrichment analysis identified significant Gene Ontology terms, including "vascular-associated smooth muscle cell apoptotic" potentially associated to the role of adipose tissue on metabolic regulation. Notably, peroxisome proliferator-activated receptor gamma (PPARG) gene was upregulated in fat tail, highlight the role of this depot in fat accumulation. However, two DEIs were more expressed in perirenal tissue, indicating nuanced regulatory mechanisms which should be further explored. Overall, our study elucidates significant differences in AS events and isoform expression between the perirenal and tail fat depots in suckling lambs, underscoring the complexity of adipose tissue regulation.

Exploiting WGCNA to analyse the suckling lamb liver transcriptome profiles of males and females

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Suckling lamb meat is one of the most relevant products of the Mediterranean dairy sheep industry. The liver, with a central role in macronutrient metabolism and homeostasis, may influence livestock carcass quality. Transcriptome sequencing, or RNA-Seq, can help us to understand the complexity of eukaryotic transcriptomes, whereas Weighted Gene Coexpression Network Analysis (WGCNA) appears as a valuable approach to unraveling the complex relationships between gene expression patterns and quantitative phenotypic traits. Thus, the present study aims to exploit the potential of WGCNA to better understand the link between liver transcriptome profiles and quantitative traits related to carcass quality in male and female suckling lambs. WGCNA identified 26 and 27 modules in males and females, respectively, and the correlation between module eigengenes and carcass traits revealed two modules in males significantly associated with the studied traits, the green module for %muscle ($r^2 = -0.88$, $p = 0.02$) and the pink module for %cavitary fat ($r^2 = -0.84$, $p = 0.04$). No significant female-related modules were identified. The green module, negatively correlated with the %muscle, was enriched in GO terms related to hypoxia and response to stress. Hypoxia has been reported to inhibit muscle growth and cell differentiation, highlighting the importance of hypoxia/stress-related processes concerning muscle. The pink module, negatively correlated with %cavity fat, was significantly enriched in GO terms related to vascularization, which is shown to be a crucial process in adipogenesis. The results here reported highlight sex-dependent differences in the liver transcriptome between sexes associated with the molecular mechanisms underlying carcass traits and underscore the importance of hypoxia and vascularization in regulating muscle and fat deposition for male suckling lambs.

The genotypes of Myostatin gene are associated with weight gain indicators of intensively fattened lambs of Latvian sheep

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MSTN, a member of the TGF β superfamily, negatively affects skeletal muscle growth and controls muscle development. Feed costs make up a significant portion of daily costs in sheep farming. Improving feed efficiency or carcass yield of lambs can reduce costs and increase income. Genetic variations of MSTN can be used in Marker association selection in animal husbandry. The study aims to search for genotypes of multi-locus of MSTN in association with weight gain indicators in Latvian sheep. Genomic DNA of 76 controlled, intensively fattened lambs from six Latvian breeds were sequenced MSTN. Variable 24 loci were identified and genotypes of multi-locus of promoter and exon 1, intron 1 and 2, and 3'UTR region were associated with fattening indicators. Nine genotypes and five haplotypes were detected based on the four SNPs in promoter and exon 1. In introns 1 and 2 were found nine and five SNPs, respectively, forming 14 and 10 genotypes, and 9 and 8 haplotypes, but in 3'UTR were four variable loci forming seven genotypes and four haplotypes. SNPs from the 3'UTR region were significantly associated with lamb birth weights, dry matter intake, average daily weight gain during feeding, and percentage of carcass weight to live weight of lambs. Genotypes from intron 1 were found in association with the percentage of carcass weight. Our results indicate that MSTN SNPs in the intron 1 and 3'UTR regions are important for intensive lamb fattening. Their use in breeding can improve Latvian sheep breeds and economic yield. Acknowledgements. The study was funded by the LZP-2021/1-0489 project

Estimating haplotype and mutation effects in the context of genome sequence via ancestral recombination graphs

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Genomic prediction with whole-genome sequence data is challenging due to high computational cost and limited increase in accuracy but it has the opportunity to capture causal mutations and their effects in the context of the genome sequence for biological discovery. To overcome the challenges, we developed a statistical model that leverages local tree structure in an ancestral recombination graph (ARG). ARGs describe genetic variation in the sampled genomes with past mutation, recombination, and branching/coallescence events. We parameterise the difference between ancestor-descendent haplotype values by the effect of the mutation(s) between the haplotypes (=branch effects). Thus, haplotype values equal the sum of all branch effects from the tree root to the haplotype. This makes the mutation effects sequence-dependent, capturing local epistasis and ancestry-specific effects. The tree structure generates a recursive and sparse model via conditional distributions of ancestor-descendant haplotype values, allowing efficient calculations via generalised Cholesky decomposition of variance and precision matrices between haplotype values. We demonstrate the model with a sample of cattle mtDNA and associated real/simulated phenotypes. Since mtDNA is non-recombinant, we work with a single ARG spanning the entire genome. Results show how our mutation-event-aware model differs from the standard variant-allele-dosage model and generates more accurate estimates of mutation effects and haplotype values in different genetic backgrounds. Extending the model to recombining genomes still imposes a significant computational challenge and is ongoing research.

Genomic prediction using functional annotations and QTL features in dairy and beef cattle breeds

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Existing Bayesian genomic evaluation methods such as BayesRC can account for biological annotations but not more than one annotation per SNP. However, when a SNP is associated with multiple sources of functional annotation, it is not straightforward to identify the most relevant annotation for a trait of interest. To handle multi-annotated SNPs, the BayesRC π approach assigns a multi-annotated SNP to its optimal annotation and within a specific effect class (null, small, medium or strong effect). These strategies were tested in two dairy breeds (Montbéliarde and Normande) and one beef breed (Charolaise). Four different annotation classes were considered (50K chip, GWAS, GWAS meta-analysis, genomic features) for a total of around 100,000 and 50,000 SNPs in the dairy and beef breeds, respectively. The traits of interest were milk, protein, and fat yields, protein and fat contents in dairy cattle, and weight at 18 months, thickness of bones, muscular and skeletal development in beef cattle. Although assuming different variance priors in a BayesR model led to a significant improvement in the accuracy of genomic predictions for a number of traits, incorporating annotation classes via the BayesRC and BayesRC π models did not result in any additional gain. However, the posterior distribution of SNPs in the different effect classes (null, small, medium or strong) strongly differed between models, with the SNPs having a strong biological annotation being more frequently assigned to the medium and strong effect classes with the BayesRC π model than with the BayesR model. This suggests that the biological information is useful in identifying SNP with strong effects, which may favour more robust prediction equations over time.

A genome-wide association study to identify novel genomic regions associated for aviary with winter garden usage by laying hens

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The health and welfare of a laying hen in an aviary system is often expressed through the frequency of movement within the aviary. However, the usage of the zones within the aviary by the hens is not fully known, but connections between zone and production have been made. Recent studies have shown that duration spent in different zones within the aviary is heritable. Therefore, the objective of the current study is to identify genomic regions or single nucleotide polymorphisms (SNP) associated with the use of different zones by hens in the aviary using a genome-wide association study. For this study, 1,106 Dekalb white laying hens (Hendrix Genetics) were genotyped using a 60K SNP array and were monitored daily for 290 days, where each visit and duration in the different zones were recorded, culminating in 937,740 records. Hens were kept in an aviary system with 5 different zones, a top level tier, nest box tier, lower level tier, floor littered area and an outdoor winter garden area. The current study aims to further provide insight into the genetic contribution for different zone usage by laying hens, highlighting potential genes associated with the traits and allowing further understanding into selecting for these behaviour traits. Ultimately, this will help to improve the welfare of animals within the aviary by selecting potential genomic regions responsible for the usage of different zones in the aviary.

Detecting NGT Products: The Role of the GenEdit Database

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The advent of new genomic techniques (NGTs) such as genome editing has revolutionised the ability to precisely edit and change the genetic code of animals. This has stimulated the launch of European initiatives aimed at further exploring the functionality of the genes of farmed animals (genotype-to-phenotype research, such as the EuroFAANG projects), developing new breeding strategies to help ruminants adapt to climate change (RUMI-GEN), or developing innovative methods to detect NGT products (DETECTIVE). One of the tasks of the DETECTIVE project is to gather information on the proposed innovations to determine their origin, the associated traits, the most frequently modified genes, and the importance of modifications. This will help define detection strategies and develop methods to distinguish NGT products from conventional ones. For this purpose, we developed the GenEdit database, based on an open-source Shiny application, which currently gathers information on 1,158 genome-edited (GE) organisms, including 314 GE animals. Among the methods used to edit animals, the CRISPR-Cas technique is overwhelmingly dominant (86%), followed by TALEN (8%) and ZFN (4%). The 10 most modified animals, in ranked order, are pigs, cattle, sheep, goat, tilapia, chicken, rabbit, Atlantic salmon, and catfish. The most listed modifications have biomedical applications (xenotransplantation), improve resistance to diseases and to biotic stresses (mainly viruses but also bacteria), increase muscle mass (mainly linked to a modification of the myostatin gene), and modify milk composition (reduction of allergenic compounds, increase in anti-stress compounds or fatty acid content).

The Genome sequence of the Nguni Sheep, *Ovis aries*

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Nguni Sheep are indigenous to the Southern Africa region. They are common within the small holder and poor resources farmers. They are well adapted to a different agroecological regions. The genomic selection for Nguni sheep has been a challenge as there is no high-quality reference genome for African indigenous Sheep. The objective of the study is to generate long reads reference genome of the African indigenous sheep. DNA samples were collected and sequenced using PacBio HiFi and Omni C. Assembly was carried out using the vertebrate genome project pipelines which allows to generate error-free, near gapless reference-quality genome assemblies. The genome size is 2.97 gb with the homozygosity of 99.8% and heterozygosity of 0.236 % with the estimated homozygous read coverage of 32x. The BUSCO analyses had 97.4% complete BUSCOs, 1.3 % fragmented BUSCOs, 1.3 % missing BUSCOs and a total of 3354 BUSCO groups searched. The total number of scaffolds is 823 with the scaffold N50 of 96,736,211 and scaffold L50 of 9. The genome assembly will serve as a baseline information to create a full reference genome for Nguni Sheep which will allow better genomic selection practice.

Whole Genome Sequencing of Aosta cattle to assess breed genomic diversity

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The dual purpose Aosta cattle breeds are composed of the Aosta Red Pied (ARP) and the Aosta Black Pied – Chestnut, with the subgroup of Hérens ancestor's Chestnut individuals, (ABCH). This cattle breeds have been raised in the Aosta valley since the 5th century without introgression from other breeds. Their farming practices make them adapted to the harsh summer pasture mountain environment. WGS data were produced for the first time in this population providing a unique source of genomic information to map possible alleles related to their capability to cope with mountain harsh environmental conditions. For this study 45 bulls, born between 2018 and 2022, have been Illumina re-sequenced at an average coverage depth of 20X (15 bulls) to 30X (30 bulls) and aligned against the ARS-UCD1.2 cattle reference genome using BWA aligner. Variants have been called with SAMtool, BreakDancer and CNVnator software's. An average of 628,686,387 raw reads have been produced with an average mapping rate of 98.9% for each sample. On average a total of 6,085,595 SNPs with 45,904 exonic variants, 1,881,945 intronic ones and a ts/tv factor of 2.25 per sample have been found. Additionally, an average of 803,381 InDels 20,956 SVs and 8,022 CNVs per sample were identified. The exonic CNVs were 811 and 753 the intronic ones, with 2372 duplication and 5650 deletion events. Genes annotated within SV and CNVs have been classified for their function with Cytoscape software. Acknowledgements: Funded by: 1) DU-ALBREDDING – PSRN 2014/2020 CUP-J71J18000020005; 2) PRIN 2022 – Research Project n. 2022F43HWL CUP- G53D23004040006.

Genetic diversity of local chickens in the Mediterranean area: Morocco, Algeria and Tunisia

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Local genetic resources are crucial for poultry sustainability around the world. Their characterization and conservation are thus capital for poultry sector. This study aims to characterize genomic diversity of local chicken populations in three countries from North Africa. A wide-scale sampling has been done considering geographic diversity over the three countries and 368 individuals from Morocco, 286 from Algeria, and 115 from Tunisia have been considered and genomic data have been produced using IMAGE001 v2 DNA array (10K SNPs). Genetic diversity within and among location and countries was assessed through different approaches: heterozygosity, inbreeding, Fst, ancestry, PCA, and phylogenetic analysis. Heterozygosity was moderate, 0.41 in Morocco, 0.46 in Algeria, and 0.42 in Tunisia. Furthermore, Fst results showed no clear differentiation between the 3 countries. The obtained values are closely clustered, from 0.0108 between Moroccan and Algerian chickens to 0.0122 between Tunisian and Algerian chickens. While, PCA analysis showed genetic similarity. On the other hand, ancestry results showed a strong admixture, particularly evident between the populations of Morocco and Algeria. In addition, Treemix analysis aligned with previous sNMF findings, indicating diverse ancestral populations and genetic mixing events within each country, irrespective of geographical proximity. These findings suggest that neutral genomic diversity within local chickens in the Western part of North Africa is adequate and further studies in selection signatures associated to local adaptation would help conceiving management plans for conservation in this region. Keywords: Chicken, local genetic resources, Genetic diversity, Population structure, North Africa, conservation.

Session 16

Theatre 2

A genomic-based approach to redefine herd book information in two local cattle breeds

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Reggiana and Modenese are autochthonous cattle breeds of the North of Italy. Their milk is mainly used to produce breed branded Protected Designation of Origin Parmigiano-Reggiano cheeses. As in most local breeds, pedigree records in Reggiana and Modenese herd books are not always complete. In this study, we applied a few approaches to verify and complete pedigree information: opposing homozygotes (OH) that identifies parentage inconsistencies in candidate parent-offspring pairs; methods that apply Identity by Descent (IBD). Pedigree information was evaluated on a total of 3400 Reggiana and 630 Modenese cattle (which represent about 3/4th and 2/3rd of the respective breed populations), using genotyping data obtained from the GGP Bovine 150k single nucleotide polymorphism array. PLINK, and KING software and ad hoc scripts provided pairwise comparisons of the genotyped cattle. By comparing OH and IBD approaches and estimating the genotyping error rate, a threshold of inconsistent OH SNPs to correct a record and to infer up to 2nd or 3rd relationship degree was defined. These methods made it possible to correct and complete Herd Book information in the two breeds. These approaches will improve conservation strategies in Reggiana and Modenese breeds. Acknowledgements: Funded by the Programma di Sviluppo Rurale Nazionale (PSRN) Dual Breeding 2, co-funded by the European Agricultural Fund for Rural Development of the European Union and by the MASAF.

Effects of indirect genetic effects on fish breeding programs

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Indirect genetic effects (IGE) have been reported in aquaculture despite incipient applications in commercial breeding programs. Although competitive-aggressive behavior has been frequently reported in fish, there is some evidence of neutral and collaborative behavior as well. We studied the potential impact of IGE determining social behavior in French fish breeding programs, using simulation of three independent traits: one phenotypically measurable and two latent ones that modulated the social interactions (SI) between individuals and their outcome to the observed phenotype. Three scenarios of SI, competition (C-), neutral (N), and collaborative (C+), were assessed. SI occurred between individuals physically close and with probability equal to a logit of the sum of their respective responses for the latent variables. This probability was used as a parameter to sample from a Bernoulli distribution to determine whether the interaction occurred. We studied 3 selection strategies based on a partial-factorial mating plan of 200 parents crossed into 10 groups of 10 females with 10 males. Parents were selected (1) among 2000 phenotyped candidates from all mating groups (IN), (2) as the best group for the observed phenotype (G), or (3) as the less-variable phenotypic group (Gv). Selection strategy G had the highest phenotypic and genetic gain for all SI scenarios, followed by IN for scenarios N and C+, and Gv for scenario C-. G and IN had similar results across SI scenarios, while phenotypic and genetic gain decreased from C- to C+ with Gv. Information on the favorable or unfavorable SI affecting commercial traits in a population under selection is thus important for defining relevant breeding strategies.

Session 16

Theatre 4

Selection for protein digestibility in pigs

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Protein efficiency in animal production will become increasingly important. Cost of protein as a feed ingredient will increase as will the cost for N-emissions. Genetic selection for improved apparent total tract protein digestibility (ATTDP) might help in reducing feed costs and reducing the environmental impact of pig husbandry. ATTDP can be calculated applying Near Infra-Red technology (NIR) on both feed and fecal samples and is a heritable trait. In a Norwegian study a strong genetic correlation between ATTDP and feed intake from 40 to 120 kg was found (-0.54 ± 0.11), which raises the question what is the potential added value of genetic selection for ATTDP? At the central boar testing facility of Topigs Norsvin in Canada, 307 fecal samples were collected on selection candidates of a sire and dam line, prior to off-test. Together with a feed sample, these fecal samples were chemically analyzed and used to develop a calibration curve for NIR equipment to be able to define ATTDP. This calibration curve was applied on fecal samples of 1705 animals in the same facility, with an average ATTDP of 78.4%. Genetic parameters were estimated for ATTDP, DFI and FCR. Low-protein diets affect ileal amino acid digestibility in growing and finishing pigs, in such a way that animals become more efficient at limiting SID Lysine levels. A correction factor, based on estimated protein deposition and feed intake one week prior to fecal sampling, was developed. This study shows the potential of selection for increased ATTDP because it is a moderately heritable trait ($h^2 = 0.27 \pm 0.07$) with reasonable genetic variance ($\sigma^2_A = 5.97$). The genetic correlations with DFI and FCR were small (-0.15 ± 0.17 and -0.21 ± 0.19 respectively). A genetic variance of 5.97 indicates that mass selection could easily yield more than 1% improvement per generation. The low genetic correlations indicate that genetic selection on ATTDP has the potential to add genetic gain over and above selection on DFI and/or FCR.

Genetic variability for key biomarkers involved in body reserves dynamics in meat ewes

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The use and accretion of body reserves (BR) is a biological mechanism in ruminants to cope with negative energy balance and improve animals' adaptation. The aim of the present study was to understand the genetic background of BR dynamics using plasma biomarker profiles (NEFA, non-esterified fatty acids; BHB, β -hydroxybutyrate; T3, triiodothyronine; INS, insulin) in Romane ewes reared under two contrasting farming systems (FS; indoor, IND, n = 173; extensive, OUT, n = 486). Primiparous and multiparous ewes were monitored at three to five key physiological stages (PhySt: mating, M; mid-pregnancy, P; 2 weeks pre-lambing; bL; 3 weeks post-lambing; aL; weaning; W). The covariance components were estimated with the AIREML method with a repeatability animal model in the BLUPF90+ family. Parity, cohort, line for feed efficiency, litter size, and FS were fixed effects, while ewe, the permanent environment of the ewes, and residuals were random effects. Heritabilities for NEFA, BHB, T3, and INS ranged from 0.12 to 0.30, 0.07 to 0.14, 0.17 to 0.26, and 0.05 to 0.32, respectively, depending on PhySt (standard errors, SE 0.03 to 0.04). Genetic correlations between the PhySt ranged from 0.07 to 0.86, 0.41 to 0.9, 0.44 to 0.86, and -0.06 to 0.94 for NEFA, BHB, T3, and INS, respectively (SE, 0.06 to 0.14). Further analyses are in progress to estimate genetic correlations between biomarkers and identify associated QTL. In conclusion, genetic levers could be used to improve the adaptation of ruminants through BR dynamics. [Funding with iSAGE project 679302]

Genome-wide SNP markers from ddRAD reveals the population structure of Italian donkey populations

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Although more than 150 breeds and 53M donkeys (*Equus asinus*) are recognized worldwide, there is no commercial SNP array for this species. The ddRAD-seq technique was used to characterize the population structure of nine Italian donkey populations. More than 418M reads were generated and demultiplexed to obtain 1.90 million reads per sample, 60K raw markers and a filtered panel of 27K SNPs across the 30 autosomes. The outcomes highlighted the informativeness of the markers, the separation of populations based on their genetic origin or geographical proximity and showed low to moderate levels of inbreeding and admixture. The success of conservation plans was highlighted for some breeds. ROH islands mapped immune-response and local adaptation genomic regions, consistent with the rusticity and adaptability of the species. A panel of genome wide SNPs offers the possibility of comparative studies among breeds of different geographic ranges to provide insights into the origin and evolution of the species.

Comparison of milk-related traits' heritability evaluated by functional controls vs Automatic Milking Systems
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Heritability (h^2) is one of the key attribute to be studied in order to assess the potential use of a trait in genomic selection. By definition, h^2 is relative to the specific animal population in which it is evaluated and is highly influenced by the measurements of the phenotypes involved. Therefore, the technologies used to record data should be carefully selected. Milk-related traits are measured by monthly milk composition analyses (MCA) in which different phenotypes (i.e., milk yield, somatic cell count, and fat percentage) are measured and are therefore considered as a gold standard measurements. Nowadays, however, Automatic Milking Systems (AMS) are increasingly available in commercial dairy farms and can record the same traits as the functional controls on a daily basis. The measurement technology is, however, not the same. In this study we compared the h^2 evaluated on different milk-related traits measured by functional controls and AMS in 5 Holstein Friesian dairy farms in a 6.35 years' time period (18,813 observations, 1810 cows). Correlations between the same milk-related trait measured with both MCA and AMS were calculated. Lastly, pedigree-based h^2 of the studied traits from the two different strategies was evaluated (using the breedR package for R) and then compared for each couple of traits. Milk yield h^2 was similar comparing the two measurement technologies (28% AMS vs 25.3% MCA). Differently, h^2 of the other milk-related traits differed when estimated on data from the two involved technologies. The results obtained in this preliminary study confirmed the importance of the methods used in phenotypes recording.

Polygenic selection in Finnish Ayrshire cows

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Finnish Ayrshire (FAY) belongs to Nordic Red breeds. With 70,000 cows in national milk recording, FAY is the second most common dairy breed in Finland. Since 2011, genomic information has been included in the breeding value evaluation and currently, the FAY breeding program is based on genomic selection. Generation proxy selection mapping (GPSM) identifies SNP markers that are strongly associated with a given proxy, e.g., birth date and is one of the few methods that are robust to distinguish selection from the random genetic drift. We identified statistically significant allele frequency changes resulting from genomic selection in FAY females using the GPSM. There were 64,148 heifers and cows born between 2009–2020 and genotyped for 43,641 SNPs. The proxy (AGE) was calculated as the period, full years, from January 2009 to the birth month of the individual. Proportion of variance (PVE) in AGE explained by genome-wide SNPs was estimated by univariate variance component estimation with AGE as the dependent variable. PVE was calculated as, where and are random polygenic and residual effects, respectively. In addition, to detect SNPs with significant changes in allele frequency over time, we conducted a univariate genome-wide association of AGE. PVE in AGE explained by genome-wide SNPs was 0.78 (SE 0.003) and was significantly greater than 0 ($P < 0.001$). There were 54 significant SNPs that have been under selection in FAY females. The SNPs were located across all autosomal chromosomes, excluding chromosomes 4, 5, and 15. Certain SNPs were located at genomic regions where selection signatures have previously been identified in FAY. The results of this study may be used to validate effects of genomic selection in FAY.

Phenotype-Genotype Linkage through Modern Data Logging: Developing a Protocol Application for Animal Research

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Objectives in animal breeding are increasingly moving away from absolute performance traits towards lifetime performance and animal health. It has been found that breeding purely for performance is usually negatively correlated with relevant health traits. In addition, there is growing recognition of the value of locally adapted breeds and small populations because they evolved in these regions and have been bred there. The agricultural use of these populations is very interesting. There is currently no freely available digital tool for livestock owners to efficiently record and manage data to scientifically prove the aforementioned advantages. The free Animal-Record application, available for Android smartphones is programmed to be backwards-compatible with smartphones used worldwide. Users can create accounts and farms, add multiple breeds and animals, and record various actions and information for each animal. For ease of use, animals can be recognized using the barcode on the ear tag or by NFC code. Furthermore, it includes an offline mode for areas with limited network coverage; data can be entered without internet connection and is then synchronized later. The secure server of the University of Kassel hosts the phenotypic data, linked with available genotypic data, enabling the calculation of animal breeding figures which can be displayed in the app on request. Further research is needed to examine the advantages and disadvantages of locally well-adapted breeds and small populations. The Animal-Record application is a digital tool that allows livestock owners to efficiently record phenotypic data. The data is stored securely and, if available, automatically linked to genotypic data. With the appropriate software, the linked data can be used for research.

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Development of an analytical tool for animal genetic progress assessment based on gEBVs and results of its on-farm implementation

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When dealing with animal genetic progress, it is essential to consider retrospective animal breeding values, genomic data, and historical herd performance. All this information allows to improve the quality of breeding decisions and provides insights into the impact of different herd management strategies. We report on the development and use of an advanced herd genetic progress analytical tool for dairy cattle that have been tested on farms for three years. Our report shows how such analytical approaches improve breeding processes and overall genetic gain for the farms that used them. We also overview how the tool utilizes genomic breeding values calculated with ss-GBLUP model, pedigrees and genotypes to build retrospective herd genetic progress reports, such as different breeding groups analysis and comparison and analytics of used bulls within a herd. As well as show an approach to calculate different herd development scenarios based on retrospective breeding data and how future herd genetic gain will change depending on different breeding strategies utilized. This allows farmers to gain insights into herd management processes such as cattle sales and purchase impact, animal culling impact, and the effect of used bulls on the specific herd or the entire population. Results of an on-farm implementation showed that this analytics enables farmers to identify weak herd management practices and improve decision-making processes in dairy cattle breeding to obtain better genetic gain.

Beef-on-Dairy leads to increased longevity of Holstein cows

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Studies show a strong genetic progress in longevity of German Holsteins, however the phenotypic trend is only moderate. This discrepancy between phenotypic and genetic longevity is often explained by management decisions, such as rearing all female dairy calves, which are later used for the replacement of older cows. Consequently, high replacement rates are resulting in early culls of cows. Since 2017, farmers have increasingly been using beef semen on dairy cows (Beef-on-Dairy, BoD). As a result, the number of female dairy calves for replacement is reduced and cows stay longer in the herd. The phenotypic longevity should then be higher in herds with less female dairy calves. To proof this hypothesis, over 18,000 herds in Germany were clustered into groups based on the proportion of female dairy calves born in 2020/2021. The average longevity of cows culled in 2022/2023 (replaced by calves born 2020/2021) was compared. There was no difference in genetic level (EBV longevity) between the groups of farms. Herds with < 40% female dairy calves (low number of replacement heifers) had an average longevity of 1330 days, whereas herds with > 55% (high number of replacement heifers) had lower average longevity (1193 days). This difference of approx. 150 days longevity indicates that many cows are culled early due to management decisions rather than for health reasons. The increasing use of BoD is one of the main reasons for less replacement heifers in herds and thus drives farmers to keep their cows longer. The achieved genetic potential is mainly limited by management and could result in higher phenotypic longevity when cows are given the chance to stay longer in a herd.

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Assessing genome complementarity in beef-on-dairy crosses through selection scans, runs of homozygosity and genomic variation

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Crossing dairy cows with beef breeds can improve offspring performance due to increased meat yield and heterosis-related fitness gains. However, crossing might also diminish traits that were previously selected in the dairy breed, such as adaptation to local environments. To assess genome complementarity in crossbreds, we use low-pass sequencing data of 181 Brown Swiss (BSW) dairy cows, 34 Angus, Limousin and Simmental beef bulls, and 301 of their F1 crosses. For each beef breed, we compare genome-wide patterns of runs of homozygosity (ROHs) and genetic variation at loci putatively under selection in BSW, and test association of the identified candidate mutations with several growth- and carcass-related phenotypes in F1s. While some islands of long ROHs remain at common and breed-specific genomic positions in F1s, suggesting shared selection pressures between beef breeds and BSW, genome-wide ROHs are substantially reduced in F1s compared to BSW. Nevertheless, very few candidate genes under selection in BSW are nearly fixed for coding genetic variation in BSW and simultaneously have high frequencies of alternate variants in the investigated beef sires, as expected with BSW-specific selection pressures. Dosages of beef-specific alleles at these genes affect economically important traits. As all alleles remain polymorphic across beef breeds, genotype-informed selection of mating pairs may be used to modulate desired phenotypes. Together, our results suggest that fattening performances of beef-on-dairy cattle can be optimized through the selection of beef bulls without affecting genomic composition at the majority of candidate genes under selection in BSW, by simultaneously decreasing the potentially negative effects of past inbreeding.

Milk yield, weight gain, feed intake, and feed efficiency in second lactation Holstein x Red Dairy Cattle F1 crossbreds compared to Holstein and Red Dairy Cattle

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Holstein (HOL), Red Dairy Cattle (RDC) and F1 crossbreds (F1) at the Danish Cattle research Center (DKC) fed the same total mixed ration were compared for feed efficiency (FE). Data originated from 12 HOL, 9 RDC and 12 HOL x RDC F1 crossbred 2nd parity cows. The cows were born in August and September 2019 and have been observed in the crossbreeding experiment at DKC since the age of one month. All cows had information on daily feed intake, body weight and milk yield. Monthly recording of fat and protein percentages were used to calculate daily energy-corrected milk (ECM), protein, and fat yield from 5 to 180 DIM. Because not all cows had consecutive observations for 180 DIM, the data was grouped in three periods: 5-60 DIM, 61-120 DIM and 121-180 DIM. 10 HOL, 8 RDC and 8 F1 had data up to 180 DIM. SAS GLIMMIX were used to predict daily ECM and milk yield, body weight, and dry matter intake (DMI). Feed efficiency was calculated as ECM/DMI ratio and protein efficiency (PE) as (kg protein yield + kg of protein in body weight change)/DMI ratio. Holstein had the highest ECM yield and DMI in all three periods and RDC the lowest. Heterosis for ECM for F1 ranged between 3% (5-61 DIM) and -2% (121-180 DIM), and for DMI -3% (5-61 DIM) and 3% (61-120 DIM). In all three periods, F1 had the highest average daily gain (ADG). F1 had the highest FE in the first period with a heterosis of 5%, while heterosis was -1% in the 2nd and 3rd period. PE was highest for F1 in all three periods (0.12, 0.22 and 0.32). During the first 180 DIM, F1 had a higher PE and ADG compared to HOL. In the first 5-61 DIM, F1 had the highest FE while HOL had the highest FE during 121-180 DIM. This indicating that that the F1 tend to utilize the food to weight gain to a greater extent than the purebreds do.

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Performance of purebred dairy cows and crossbreds between Swedish Red, Swedish Holstein, Jersey, and Montbéliarde in Swedish herds

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This study aimed to compare crosses between Swedish Red (R), Holstein (H), Jersey (J), and Montbéliarde (M) to purebred R, H, and J for fertility, calving, production, and survival traits. The main focus was on F1 crosses between either H or R on one hand and J or M on the other hand, but also 3-breed crosses were studied. There were 2,154,241 observations for cows that calved between 2005 and 2020. The data set was separated into two parity groups (1st and 2nd) and analysed by using a mixed linear model. Fertility interval traits and number of inseminations were improved in the F1 crosses with J compared with pure R or H, more clearly so in the first parity. In parity 2 the difference was sometimes not significant. The significant improvements of fertility interval traits in parity 1 for F1 crosses with M compared with pure R or H were generally smaller than for crosses with J. The results for calving difficulty and stillbirth for Jersey crosses were clearly influenced by the levels for the purebred J. Because J was better than R and especially H for calving difficulty, F1 crosses with J were also better than R and H, respectively, but not significantly better than pure J. J had the highest stillbirth rate so crosses with R and H generally significantly decreased stillbirth rates compared with J, however, not to the extent to make F1 crosses better than purebred R or H. For M, F1 crosses were not better than pure R or H for calving difficulty or stillbirths. F1 between J and H (JH) had higher 305-day fat yield than H, but lower milk and protein yields. MH had higher fat and protein yields than H and MR had higher 305-d milk and protein yields than R.

Heritability and Genetic Correlations of Resilience Indicators in Beef Cattle

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This study aims to estimate heritability and genetic correlations for standardized weight at 550 days of age (P550) and resilience indicators related to environmental challenges during the growth phase in Nelore heifers. Data from the Institute of Animal Science belonging to three selection lines were used: NeC, under stabilizing selection for yearling weight (YW); NeS, selected by the highest selection differential for YW and NeT, selected by the highest YW and smallest RFI. The dataset comprised 2,871 heifers and 18,783 weighing records from birth to YW (565 ± 50 days of age). Different growth models were accessed including repeated measures, Brody, logistic and orthogonal Legendre polynomials. The residues were used to calculate resilience indicators: natural logarithm of variance (LnVar), autocorrelation (rauto) and skewness. The BLUPF90+ program was used. The model included fixed effects of selection line (NeC, NeS, NeT), year (1981-2019) and month (9-11) of birth, cow age as a covariate and genetic additive as a random effect. The repeated measures model was the best to described growth. W550, LnVar, rauto and skewness presented h^2 of 0.48 ± 0.03 , 0.38 ± 0.04 , 0.12 ± 0.03 and 0.10 ± 0.03 , respectively. The rg were 0.52 ± 0.06 (P550 x LnVar), -0.23 ± 0.11 (P550 x rauto) and 0.55 ± 0.12 (P550 x skewness). The h^2 of resilience indicators revealed sufficient genetic variability for inclusion in genetic improvement programs. The findings demonstrate the importance of incorporating resilience indicators into breeding strategies to enhance productivity of beef cattle.

Session 16

Poster 16

Genomic-based genetic parameters for resilience indicators and pregnancy in beef cattle

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This study aimed to estimate genetic parameters for pregnancy (PREG) and resilience indicators associated with environmental disturbances in Nelore heifers. Animals from three selection lines were utilized: NeC, under stabilizing selection for yearling weight (YW); NeS, selected by highest selection differential for YW; and NeT, selected by highest differential for YW and lower RFI. Data included 3,072 heifers and 24,548 weight records (WR) from birth to first breeding season (742 ± 68 days of age). Growth was estimated with sixth-order Legendre orthogonal polynomials model. Residuals were used to calculate resilience indicators: natural logarithm of variance (LnVar), autocorrelation (rauto), and skewness. Genotypes from 3,226 animals and 383,856 SNPs were used. Gibbs sampling was used to estimate h^2 and rg with GIBBSF90 and POSTGIBBSF90 software. The analysis included 1,500,000 iterations, burn-in of 150,000 and thinning of 10, considering a threshold model for PREG. Systematic effects were selection line (NeC, NeS and NeT), year (1981-2020) and month (9-11) of birth, cow's age as covariate and genetic additive as random effect. The h^2 for PREG, LnVar, rauto and skewness were 0.07 ± 0.04 , 0.42 ± 0.03 , 0.29 ± 0.03 and 0.10 ± 0.02 , respectively. The rg were -0.26 ± 0.09 (PREG x LnVar), -0.24 ± 0.09 (PREG x rauto) and -0.16 ± 0.10 (PREG x skewness). The negative genetic correlations between PREG and all resilient indicators suggest that heifers with less impact (LnVar), fast recovery rate (rauto), and null influence (skewness) of environmental disturbances have a higher probability of becoming pregnant.

Estimation of genomic inbreeding coefficients based on SNP markers in Serbian Holstein cows

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In the traditionally breeding schemes inbreeding is calculated based on genealogy data, and the accuracy of the estimates depends on the depth and reliability of the data in the pedigree. With the availability of SNP markers, it became possible to calculate genome based inbreeding coefficient with the aim of determining homozygosity on markers. In order to estimation of genomic inbreeding coefficients, a total of 1,600 Serbian Holstein cows were genotyped with a GGP Bovine 100K SNP array. Data quality control showed that 1,591 cows had a call rate above 90%, while 3,958 SNPs had a minor allele frequency below 0.01, which is why they were excluded from further analysis. Individual inbreeding was calculated using the `-het` function within the PLINK software package. Among 629 (39,5%) cows, the presence of identical markers from both the paternal and maternal sides was determined, indicating inbreeding, which had a maximum value of $F_x=0.223$, while the average value for this number of individuals was $F_x=0.023$. Additional research and genotyping of a larger number of cows are needed in order to estimate population inbreeding. Acknowledgments: This research is funded by Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Contract No. 451-03-65/2024-03/200117). The data used in this research were collected during the realization of PROMIS project: 'A Bioinformatics Approach to Dairy Cattle Breeding Using Genomic Selection', No. 6066512, funded by the Science Fund of the Republic of Serbia.

CapraGEN project: managing genetic variability in Alpine and Saanen goats through genomic approach

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The economic importance of goats in European agriculture is rising due to increased demand for dairy products. Italy is acknowledged as a repository of genetic resources for caprine species, with 37 breeds as documented by National Goat and Sheep Breeder Association. CapraGEN project, titled "Application of genomics in dairy goat farming", aims to understand variability within flocks and the genetic relationships of goats farmed in Lombardy region. A total of 950 Alpine and Saanen goats were sampled and genotyped using the GeneSeek Genomic Profiler (GGP) Goat 70k. Principal component analysis (PCA) illustrates the graphical representation of how individuals and flocks cluster based on their genotype. Among the genomic tools and methodologies aimed at characterizing animal biodiversity, the examination of runs of homozygosity (ROH) stands out as one of the most valuable approaches. ROH length, ROH islands and FROH are indeed proposed as useful markers for estimating the timing of the associated inbreeding event. In this study we analyzed ROH length, their distribution, the presence of ROH island and the derived inbreeding values (FROH) in 950 goats. A total of 32,089 ROH were identified with an average of 33.81 ROH per animal. The average FROH calculated for ROH longer than 16Mb was 0.021 for Alpine and 0.029 for Saanen, suggesting a recent inbreeding event in both breeds. For both breeds, we assessed the presence of ROH islands, defined as genomic regions containing ROH of varying lengths. Within these regions, Alpine goats exhibited SNPs shared by at least 25% of the population on chromosome 6. In Saanen goats, peaks on chromosome 11 were identified, but shared by only 15% of the population. The analysis of ROH distribution within each flock, the presence of ROH islands, together with the level of inbreeding and evolution across generations revealed the presence of different management practices. Funded by EAFRD Rural Development Program 2014–2020, Management Authority Regione Lombardia – OP. 16.1.01, Project ID n. 202202380565 – 'Operational Group EIP AGRI'; <https://ec.europa.eu/eip/agriculture/en/eip-agriprojects/projects/operational-groups>.

Genome-wide scan for lethal haplotypes in Nelore cattle

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The genome-wide scan has enabled the identification of lethal recessive alleles that are the major genetic causes of harmful effects on production traits. Hence, this study aims to identify lethal haplotypes based on the absence of homozygous haplotypes in Nelore cattle. A total of 2,446 animals were genotyped with medium density assay (GGP 50K and 75K SNPs), and 780 animals were genotyped with Illumina BovineHD panel (770K SNPs). Animals genotyped with medium density were imputed to the HP panel using FImpute v.3 considering the ARS-UCD1.2 Bos Taurus genome assembly, and a total of 612,154 autosomal SNP markers remained for analysis. The haplotypes were constructed using the sliding windows method implemented in findhap.f90 software and was classified as lethal when attending the following criteria: haplotype frequency higher than 2%, expected number of homozygous carriers greater than 1, and probability of observing 0 homozygous carriers lower than 0.6. The ENSEMBL software was used to search for genes harboring the regions of lethal haplotypes. A total of 45 genomic regions were identified as potential candidates for lethal haplotypes. The haplotype with the highest frequencies of heterozygous carriers and 0 homozygous was located on BTA15, with a frequency of 11.39% and 735 carriers (22.04 – 22.78 Mb). The lethal haplotypes regions harbored candidate genes related to the major functions of the immune system, metabolic functions related to energy homeostasis, and embryonic development. Identifying these genomic variants is essential for designing mating strategies and avoiding the spread of lethal alleles in the Nelore population. Funding: FAPESP (#2017/10630-2 and #2023/11176-4) and CAPES (Finance Code 001).

AgriseqPI 1.0 & AgriSeqRI 1.0: Reporting utilities for SNP based parentage determination and testing for traits and disorders with Targeted Genotyping by Sequencing panels

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The AgriSeqPI is a bulk report generation plugin primarily developed for SNP based parentage and identity analysis using the AgriSeq™ Targeted Genotyping by Sequencing (T-GBS) parentage panels for canine, feline, bovine, equine, ovine, caprine run using the Torrent Suite™ Software (TSS). The reporting plugin is based on the ISAG rules and guidelines for parentage determination using SNP markers. End users can generate individual reports for each sample on a sequencing run using AgriSeqPI. Similarly, The AgriSeqRI is a bulk report generation plugin primarily developed for the AgriSeq™ Traits and Disorders panels for canine, feline, bovine, equine, ovine, caprine run using the Torrent Suite™ Software (TSS). End users can generate individual reports for each sample on a sequencing run using AgriSeqRI. AgriSeqPI and AgriSeqRI can be run manually or automatically via plan run after a sequencing run is completed on Ion Torrent. Both plugins retrieves the genotype data from Torrent Variant Caller (TVC) and AgriSum Toolkit and outputs a comprehensive report for each sample for all markers in each panel. The AgriSeqPI report adheres to the ISAG parentage determination guidelines and includes sample, reference, panel (test) details, and a summary of the results. The AgriSeqRI outputs a comprehensive report for all genetic disorders and associated trait markers for each species. The report includes sample, reference, panel (test) details, and a summary of the results. In addition, information including the condition, genotype (reference and variant allele), inheritance type, and OMIA reference, is provided for each marker. A brief explanation of the genetic trait, breed specificity, genetics and inheritance gathered from several publicly available publications are also reported. For Research Use Only. Not for use in diagnostic procedures.

Resilience indicators in beef cattle selected for high performance

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This study aims to evaluate the effects of selecting for yearling weight (YW) on resilience capacity of Nellore heifers in response to environmental disturbances. Data from Institute of Animal Science selection lines were used: Control Nellore (NeC), selected by zero-selection differential for YW, Selection Nellore (NeS), selected by the highest selection differential for YW and Traditional Nellore (NeT) selected by higher YW and lower RFI. Heifers were weighted from birth to yearling (565±50 days), resulting in 18,783 weight records from 2,871 animals. Five growth models were fitted: repeated measures, Brody, logistic and fifth- and sixth-order Legendre orthogonal polynomials. Weight deviations were used to calculate resilience indicators: natural logarithm of variance (LnVar), autocorrelation (rauto), and skewness. Adjusted means were compared by Tukey test considering fixed effects of selection line (NeC, NeS, or NeT), year (1981-2019), and month (9-11) of birth. Repeated measures model was the best for describing growth. NeC showed the lowest mean for LnVar at 5.69±0.03, while NeS and NeT had 6.09±0.02 and 6.12±0.02, respectively (P<0.05). NeS and NeT had mean of 0.04±0.01 for rauto and NeC 0.07±0.01 (P<0.05). Skewness means were -0.08±0.02, 0.06±0.02 and 0.14±0.02, for NeC, NeS and NeT, respectively (P<0.05). Nellore heifers selected for higher YW are more susceptible to environmental disturbances (higher LnVar). However, demonstrate fast recovery (lower rauto), and experience less negative impact from environment (positive skewness). Selection for growth traits may have implications for resilience of *Bos indicus* heifers.

Session 16

Poster 22

Genetic background of beef-on-dairy calving ease

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A recent practice in dairy herds is to breed females not selected as replacement heifers to beef bulls. This increases the market value of the surplus calves sold for beef purposes. Some beef breed associations have built selection indices for that, which include mainly carcass traits. Calving ease (CE) is also an important trait, given that some beef breeds produce high-birth-weight calves. This study investigated the genetic background of CE for beef bulls bred to dairy cows. We analyzed 1.2 million CE records (easy=1 and difficult=2) in the first three lactations from Holstein and Jersey cows inseminated with Angus, Charolais, or Simmental semen. We compared single-trait linear and threshold models with age of the cow as a covariable; sex of the calf, dam-sire breed interaction, and year-season as fixed effects; and herd-year, animal, and maternal effects as random. Calving difficulty incidences (%) were 12.2 (7.9), 11.9 (5.7), and 13.1 (16.2) for Holsteins (Jerseys) inseminated with Angus, Charolais, and Simmental bulls. Direct and maternal heritabilities for the linear (threshold) model were equal to 0.014±0.003 (0.030±0.004) and 0.19±0.003 (0.25±0.005), respectively. Maternal heritabilities were greater than direct in both models and were more similar to those found in beef breeds than dairy breeds. Linear or threshold models may provide similar EBV ranking. Although direct heritabilities were low, there is genetic variability for calving ease, and accounting for this trait when selecting beef bulls can help reduce the incidence of difficult calving when they are bred to dairy cows.

The use of cooperative herds in the promotion of genetic resources of cold-blooded horses in Poland
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Genetic resources conservation programmes for cold-blooded horses of the Sokolski and Sztumski types have been implemented since 2008. In 2023, after 15 years, the number of mares increased from the initial 228 mares to 1801 in the Sztumski type, and from 339 mares to 1468 in the Sokolski type. Due to the increasing number of horses, to preserve the environment's influence on the breed's development, and traditional utilisation of cold-blooded horses, in 2023 a change was introduced to the conservation program. For herds located outside the historical region, the term "cooperative herd" was introduced, and the activities that promote the use of horses by the provisions of the conservation program have been obligated. The following were desirable: working use in agricultural and forestry works, promotion through participation in regional events, milk production and processing, use as a leisure horse, hippotherapy, and agritourism. In total, 35 herds provided documentation confirming the different utilisation of horses. The most significant number of herds, as many as 18, use horses for work in agriculture or forestry, 17 herds take part in regional events, 2 herds produce milk from mares, and 8 herds provide hippotherapy, horse riding, or agritourism. Some of the herds (11) carry out several types of use. Cold-blooded horses, participating in cultural events such as harvest festivals, are integral to the local tradition. Hippotherapy, horseback riding, and agritourism can provide opportunities for business growth and employment for young people. This promotes the sustainable use of genetic resources and raises awareness among people about the importance of the conservation of animal genetic resources

Genome-wide association study for milk and blood indicators of hyperketonemia in Holstein dairy cattle
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At the onset of lactation, specialized dairy cows commonly experience negative energy balance that often results into hyperketonemia (HYP) or even clinical ketosis. HYP diagnosis relies on the blood level of β -hydroxybutyrate (BHB), which is known to be the major and most stable ketone body in cows' biological fluids, including milk. Recently, in fact, mid-infrared (MIR) spectroscopy prediction models for blood BHB concentration from milk spectra have been developed. Although characterized by moderate accuracy, MIR-predicted blood BHB is useful for selective breeding. We identified genomic regions associated with milk BHB and MIR-predicted blood BHB in Italian Holstein cattle to disclose potential overlaps and divergences. A single-step genome-wide association study was performed using the 2 phenotypes after log₁₀-transformation collected in lactating cows within the 35th day in milk. The 8,277 cows with phenotypes available were genotyped with various SNP panels, so data were imputed up to approximately 70,000 SNP. After conventional quality controls, 63,408 markers located in autosomes (BTA) were left. The blood BHB significant regions were located in BTA 6, 11, 14, and 25, while those of milk BHB were in BTA 11. For blood BHB, the signals with the highest P-value were in the gene PAEP (BTA 11) and DGAT1 (BTA 14). For milk BHB, the most significant signal was found within the gene FNBPI (BTA 11). Our results provide new information about the underlying biology and molecular mechanisms associated with HYP in Holstein cows.

An association study with dairy traits confirms the importance of three SNPs for the assisted selection in the Mediterranean river buffalo

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The search for DNA polymorphisms enhancing dairy animal genetics spans over 40 years, notably in cattle, identifying key alleles in casein and DGAT1 genes for milk traits improvement. Despite advanced genomic tools and precise phenotypic records, similar signs of progress have not been made in river buffaloes yet. This study aimed to explore the effects of four SNPs in CSN1S1 (AJ005430:c.578C>T), CSN3 (HQ677596:c.536C>T), SCD (FM876222:g.133A>C), and LPL (AWWX01438720.1:g14229A>G) genes on six milk traits (daily milk yield, dMY; protein yield, dPY; fat yields, dFY; fat %, dFP; and protein %, dPP; somatic cell score, SCS; and urea, MU) in a representative buffalo population. Genotyping was conducted on 800 buffaloes, and milk traits were recorded monthly from 2010 to 2021. Associations between SNPs and milk traits were analyzed using 15,742 milk test day records from 680 buffalo cows. Three out of four SNPs showed associations with various traits. CSN1S1 was associated with increased dMY ($p=0.022$), dPY ($p=0.014$), dFY ($p=0.029$), and SCS ($p=0.032$). CSN3 positively correlated with SCS ($p=0.005$) and milk urea ($p=0.04$). LPL demonstrated favourable effects on dMY ($p=0.028$), dFP ($p=0.027$) and dPP ($p=0.050$). Conversely, SCD showed no associations. This study represents the first confirmation of the importance of these genes in Mediterranean river buffalo breeding programs, providing valuable insights for preselecting young bulls for breeding.

Session 16

Poster 26

Exploring the resilience of an intensive Holstein farm in Lombardy, Italy

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The resilience of farm animals is a key to maintain productivity, longevity, and welfare, thereby ensuring the economic profitability and sustainability of livestock production. This is particularly pertinent in highly productive breeds like Friesian Holsteins. Longitudinal data on physiological traits like milk yield, feeding, and rumination time can serve as proxies for inferring resilience in farmed cows. In this study, we investigated resilience indicators in 848 cows milked in an intensive farm equipped with 9 automatic milking systems (AMS). A total of 1153 lactation curves were analyzed, with 848 first parity cows, and 533, 252, and 78 second, third, and fourth parity cows, respectively, resulting in a total of 434,679 observations. For each animal and lactation, residuals of daily milk yield were calculated after determining the best-fitting model. These residuals were then used to compute two resilience indicators: the natural logarithm-transformed variance (LnVar) and lag-1 autocorrelation (rauto) of daily yield residuals. The extreme phenotypes identified on the basis of these indicators were cross-validated with somatic cell count (SCC) data and pharmacological treatments provided by the farmer. The heritabilities of LnVar and rauto were estimated as well as the genetic correlations of these indicators with traits included in the selection indexes. This research sheds light on resilience dynamics in herds and underlines the potential of longitudinal data in the assessment of animal welfare and productivity under different environmental stressors. This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

Exploring the sustainability aspects of innovative livestock systems in Europe

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In pursuit of increased sustainability, the current European animal farming sector presents a growing number of diverse and innovative livestock systems applying specific types of innovation. Our aim was to provide a sustainability assessment (SA) of these systems to highlight common elements, strengths and weaknesses, and to provide policy recommendations for the promotion of a more sustainable European livestock sector. The SA was performed with the Public Goods tool, a multi-level assessment tool adapted within the PATHWAYS Horizon 2020 Project. Data was retrieved from 106 livestock farms specialized in different animal species (dairy and beef cattle, pigs, poultry), located in nine different EU nations and distributed in 13 national groups, practice hubs (PHs). Each PH was organized around a theme of innovation in practice. The study resulted in composite indicators of performance in each dimension for each PH. Correlations among the indicators were evaluated, and the performances of the farms compared. Environmental indicators correlated in a predominant positive way with social ones, but presented a higher amount of negative correlations with the economic indicators. Monogastric farms tended to perform similarly, whereas ruminants' farms had a wider spread. The study provides an overview of the relationships between various aspects of sustainability and a present-day frame of some of the most advanced European realities.

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Theatre 2

Adopting low carbon practices: a cost-effective strategy for French dairy farms

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Dairy farming is facing major environmental challenges, and specifically its impact on climate change. The uptake of mitigation actions is not easy and needs to be motivated. One argument is the possible positive link between economy and low carbon practices. To verify this hypothesis the Carbon€co project aims to cross analyze economics and environmental farms data from two French databases: CAP'2ER® and COUPROD®. CAP'2ER® tool evaluates environmental performances while the COURPOD® tool calculates production costs. 232 farms were identified with both an environmental evaluation and a production cost for the same year (from 2017 to 2022). The cross database was ranked according to greenhouse gases (GHG) emissions and divided into 3 classes (with significant difference): average (1,05kg eqCO₂/L), 1st decile (0,83 kg eqCO₂/L) and last decile (1,36kg eqCO₂/L). Results showed differences of economic indicators between the average and the 1st and last decile. The feed system cost was cheaper between 1st decile and average (difference of 26€/1000L). Differences are explained firstly by mechanization cost, secondly by grass and forages production costs, and lastly by the feed cost. Technical data show a better efficiency between input and output. This study confirms that environmental efficient dairy farms have also improved economical results. This can be used to engage more dairy farmers within carbon emission reducing projects.

Life Cycle Assessment of different poultry production systems around Europe: mEATquality project
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One of the main objectives of the H2020 mEATquality project is to generate information about producing more sustainably through extensive husbandry practices, meeting animal welfare and environmental concerns. The main tool used for the environmental sustainability of the farms evaluated is Life Cycle Assessment (LCA). This LCA study collects data from 60 fattening broilers farms from 2 countries (Germany, and Poland) and covers diverse models and husbandry practices, ranging from large intensive farms to small, organic, and free-range ones. The manure handling emissions were calculated using Tier 2 equations from IPPC guidelines. The not differentiable emissions, such as energy consumption, were assessed by economic approach, and environmental impacts were calculated with AGRIBALYSE® database. The limit of the system are “cradle to gate”. The carbon footprint (CF) obtained for 42 days old broilers from conventional systems was 1.40 ± 0.02 kgCO₂eq kg live weight⁻¹, with feeding representing 88% of emissions; while in organic systems broilers achieved 80 days of live with a CF of 2.67 ± 0.94 kgCO₂eq kg live weight⁻¹, and the feeding representing 47% of emissions. Besides that, different CFs of those systems, productive performances, levels of extensification and handling practices show different environmental impacts within farms and allow to differentiate systems and farms. This project has received funding from the European Union’s Horizon 2020 research and innovation program under Grant Agreement No 101000344.

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Theatre 4

Life cycle assessment of two common dairy cattle farming systems in an Alpine area
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Recently, ruminants and especially dairy farming is increasingly criticized for being inefficient in terms of environmental sustainability and because of its GHG emissions. Consequently, the aim of this study was to assess the environmental impact of two dairy farming systems, both commonly found in the alpine province of South Tyrol, Italy, applying LCA methodology. An intensively managed production system is compared with a forage-based production system. Overall, the environmental impacts were quantified focussing on four different impact categories: Global Warming Potential (GWP100), Acidification Potential (AP), Marine Eutrophication Potential (MEP), and Land Use (LU, m₂ and Pt, with the latter additionally considering the Soil Quality Index). Besides being attributed to one kg of milk (fat and protein corrected milk, FPCM), these impact categories were also related to one m₂ of farm area. Furthermore, the net protein (NP) provision was calculated for both farming systems, to provide a broader context of dairy farming, in terms of food supply and food-feed competition. The results provide an evaluation of different environmental aspects of two diverse, yet commonly found alpine dairy farming systems with the aim to illuminate the environmental sustainability in dairy farming within a broader context and to increase at the same time the societal acceptance of such livestock systems.

Quick sustainability scan calculator for intensive and extensive broiler farms

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One of the objectives of the H2020 mEATquality project is to support the development of techniques to assess the sustainability of broiler production; aiming to improve it by implementing the best practices while assessing its environmental, social, and economic impacts. Thus, two quick sustainability scan calculators were developed with those aims: one for intensive and one for extensive broiler farms. The calculators are based on the evaluation of 10 aspects related to poultry management: 1) certifications; 2) water management; 3) feed; 4) waste and residues management; 5) energy efficiency; 6) socio-economic contribution to the territory; 7) farm associated businesses; 8) animal handling; 9) management of pastures, soil and biodiversity; 10) stocking rate. The last two aspects evaluated only in extensive farms. Each of these aspects receives a score according to its importance and contributes in a different proportion to a final score (with a maximum of 100 points), which is organized into 3 categories: environmental, social, and economic impact. These calculators are designed for easy use (answers are just yes or no), with 61 and 53 questions for extensive and intensive farms, respectively. These calculators were used to evaluate 60 farms from Poland and Germany. Intensive farms achieved average scores of 81.6, 70.8, and 79.6 points in the environmental, social, and economic impact categories, respectively; while extensive farms achieved average scores of 79.3, 72.0, and 78.0 points in the same categories. These calculators are useful as a self-assessment tool for farmers, allow comparisons between farms (benchmarking), identify the weak points of each farm, enable participatory certification, and help to provide confidence to consumers. This project has received funding from the EU Horizon 2020 research and innovation program under Grant Agreement No 101000344.

Session 17

Theatre 6

A case study on carbon neutrality of Podolian beef productions in the marginal area of Southern Italy

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Podolian cattle represent a local breed, traditionally raised on pasture and woodland in southern Italy. The present study aimed in assessing the environmental impact and carbon neutrality of Podolian farms with different productive efficiency. Two farms were analysed: Podolian more efficient (PME) farm, with a total area of 200 ha, including 190 ha of permanent pasture and 10 ha of meadow hay, rearing 110 cows and 50 young bulls slaughtered at 18 months, with live weight (LW) of 550 kg and production efficiency of 138 kg of LW at slaughtered per hectare per year; Podolian less efficient (PLE) farm, with a total area of 220 ha, including 170 ha of permanent pasture and 50 ha of woodland, rearing 80 cows and 35 young bulls slaughtered at 18 months with LW of 500 kg, and production efficiency of 80 kg of LW at slaughtered per hectare per year. Carbon footprint (CF) expressed in kg CO₂-eq, was evaluated by Life Cycle Assessment (LCA) methodology, with a 100y horizontal time; the functional unit (FU) was 1 kg of LW. Carbon sequestration (CS) was also assessed. PLE farm showed higher CF than PME farm (33.8 vs 23.1 kg CO₂-eq per FU, respectively). When considering the CS on pastures, the PLE was able to reduce the CF by 75%, while the PME achieved a reduction of 72%. The PLE also achieved a higher carbon storage value and reached carbon neutrality by preserving 4.8 ha of undisturbed woodland, whereas the PME should have dedicated 4.0 ha of land to the forest in order to achieve the same benefit for carbon neutrality. The PLE farm, despite the lower efficiency, thanks to the presence of the forest, showed carbon neutrality compared to the PME farm per kg LW.

Animal Welfare Status in Dairy Cattle Farms of the Parmigiano Reggiano Consortium

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Farms within the Parmigiano Reggiano Consortium (PRC; Italy) assess cattle welfare (CW) using the ClassyFarm system. This is based on scores derived for 105 parameters by a veterinarian, these are transformed into 5 CW scores (CWS; % score) focusing on the following areas: A – Business and Personnel management, B – Structures and Equipment, C – Animal Based Measures, D-Biosecurity, and E-Total Welfare. Leveraging these data, we compared cattle welfare in free- and tie-stall housing systems in farms within the PRC. ClassyFarm CWS data, collected in 2022 from 329 tie- and 649 free stall-farms were analyzed. The average milk yield was 30.46 and 25.33 kg in the free and tie stall systems, with an average number of cows of 299 and 86, respectively. For both housing systems, CWS were above 60% (the suggested sufficient threshold). Average total welfare was 74 and 77 for the tie and free stall systems respectively. Pearson correlations among the CWS ranged between 0.15 to 0.74 (Area A – and E), with the latter highlighting the importance of area A on cattle welfare. Across all CWS areas, free stalls outperformed the tie stalls. Principal component analysis (PCA) on the CWS didn't differentiate between the two systems. However, a PCA conducted on the 105 parameter detail separated the two housing systems. Our analysis showed overall better cattle welfare status in the free housing system compared to tie stalls.

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Theatre 8

Effect of extensification factors on broiler welfare and meat quality

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We studied whether factors related to extensiveness of broiler production had a beneficial effect on welfare and meat quality. Trial 1 (T1) (Hubbard JA757 birds) comprised stocking density (SD) (21 vs. 39 kg/m²) and roughage provision (n=4 pens/treatment combination). Trial 2 (T2) comprised environmental enrichment and breed (fast, Hubbard JA987 vs. slow, Hubbard S757N) (n=5 pens/treatment combination; SD=30 kg/m²). Measurements were equal for both trials: performance, behaviour, fearfulness, play at three similar body weights; welfare indicators and meat quality (texture and colour) at slaughter. In T1, reduced SD broilers showed more foraging (P<0.05) and less resting (P<0.01); had better gait scores; were cleaner; had less footpad dermatitis and hock burn (P<0.001). Roughage had minimal effects. There were no effects on texture and colour of the meat. In T2, the slow-growing breed showed more active and foraging behaviour; had better gait, footpad dermatitis, hock burn and cleanliness scores (P<0.05). For meat texture, the slower-growing breed had higher (P<0.001) BMORS shear force values (11.0 ± 1.18 N), meaning tougher meat, than the fast-growing breed (8.5 ± 0.23 N). Color values were also affected by genetics (P<0.001). Enrichment increased play (P<0.05) and the b* (yellowness) value of the meat (P<0.05) as well as the interaction of enrichment and genetics (P<0.05) on the b* value. This implies that the breed may have a different impact on how the enrichment affects the yellowness of the meat. Thus, reducing stocking density and a slow-growing breed had most beneficial effects on welfare. However, only genetics and enrichment influence the texture and color of the meat.

Consumer expectations for beef in the French region Auvergne-Rhône-Alpes

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In France, beef consumption has been decreasing for many years. The reasons are the (i) controversial image of beef (its environmental impact, the competition between feed and food, the respect of animal welfare), (ii) the modification of consumption patterns (decrease of daily time dedicated to cooking and eating), (iii) its high price, and finally (iv) its variable sensory quality. Thus, consumers are not always satisfied with the organoleptic quality of beef. In this context, an online survey within the project OABov-AURA (2022-2024) was carried out to study consumer expectations regarding the intrinsic and extrinsic qualities of beef in the region Auvergne-Rhône-Alpes (AURA) (the 2nd largest beef cattle breeding region in France). Of those surveyed, 48% reported having reduced their meat consumption in recent years (compared to 42% who reported no change). The two main reasons for the decline in beef consumption are health (52% of responses) and ethics (44%) issues. The majority of consumers (51%) said that eating beef for pleasure was their first motivation, and are not disappointed with raw (83%) or cooked (54.5%) beef. Origin (44%), proximity (38%) and the presence of signs of quality and origin (SIQO) (34%) are the most important extrinsic criteria (% of respondents who answered “very important”). However, taste (62%), tenderness (59%) and freshness (58%) are the most important intrinsic criteria of beef for the respondents. Consumers also prefer meat that is bright red (72%) and rather fatty (64%). In conclusion, it is important that consumers in the AURA region have access to a varied supply of meat in butcher shops, of French origin and under quality signs.

Session 17

Theatre 10

Consumer preferences for cheeses derived from the milk of goats reared under different farming systems

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The study aimed at investigating the consumers' possible preferences for cheeses made with milk from goats reared under either intensive or extensive farming systems. Two types of cheeses were assessed; a semi-hard goat cheese with surface mould (cheese A) by consumers recruited in Italy, Switzerland, and the Netherlands (nA= 297), and a white goat cheese preserved in brine (cheese B) by consumers recruited in Greece (nB= 194), to record and analyze their preferences. Each panel, blind-tasted two cheeses each made by either milk derived from extensively or intensively reared goats. An overall preference for both types of cheeses produced by the milk of extensive farms was recorded (59.9% and 53.6% for cheeses A and B, respectively). Cheese A derived from the intensive farm was preferred by 25.3% (19/75), 35.6% (31/87), 49.2% (31/63) and 52.8% (38/72) of individuals aged <25, 25-40, 40-55, and >55, respectively. For cheese B, the respective values were 44.9% (44/98), 38.9% (21/54), 69.6% (16/23) and 47.4% (9/19). Regarding cheeses derived from the intensive farms, cheese A was preferred by 49.7% (76/153) of men and 30.1% of women (43/143), and cheese B by 32.0% (24/75) of men and 55.5% of women (66/119). Consumers' preferences displayed remarkable variations with regard to the cheese type, the farming system, and the consumer's gender and age. This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101000216.

Identification of breast defects in broilers with short-wave pocket near-infrared spectrometer

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Chicken breast meat is the most popular animal protein source, but three novel myopathies have been described in broilers over the last decade known as wooden breast (WB), white striping (WS) and spaghetti breast (SB). This study assessed the potential of a pocket-sized near-infrared (NIR) spectrometer to predict WB, WS and SB in broilers. A total of 4313 breasts from Ross 308 strain chicks were scanned with the pocket-sized NIR device from 740 to 1070 nm. Each spectrum was matched with the defects information, and a partial least square discriminant analysis was conducted with external validation. Results revealed that breast defects reduced the sample's absorbance. Neither the spectrum first derivative nor wavelength selection improved the models' accuracy compared to the raw spectrum. The highest balanced accuracy was obtained for detecting WS (0.80), followed by WB (0.75) and SB (0.51). All models achieved remarkably high sensitivity (>0.89), indicating their proficiency at identifying samples without defects. On the other hand, the specificity was low (<0.64). In conclusion, models developed using raw absorbance can be used without any additional mathematical treatments to identify samples without defects. Further studies are suggested to confirm if positive samples are genuinely affected by a myopathy. Funded EU H2020 101000250-INTAQT.

Session 17

Theatre 12

Does breeding for A2 cows alter milk yield and composition?

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This study aimed to investigate the effect of β -Casein (CN) family variants A1 and A2 along with κ -CN (AA, AB, AE, BB, BE, and EE) and β -lactoglobulin (β -LG; AA, AB, and BB) genotypes on milk yield (MY) and composition (protein, fat, and SCC) in Holstein Friesian cows. Data on monthly controls were retrieved from 2016 to 2022 from 6 farms. The final dataset included 23,088 observations from 1,233 cows, and values beyond the mean \pm 3 standard deviations were considered outliers. Data were analyzed using PROC MIXED in SAS ver. 9.4. Fixed effects were β -CN, κ -CN, and β -LG genotypes, farm, days in milk (DIM), parity, calving season, calving year, and the interaction of β -CN with all fixed effects except for κ -CN and β -LG. Animal nested within farm was considered as random effect. Results indicated that farm, DIM, parity, season, and year of calving affected all milk traits ($P < 0.05$) except SCC between farms. Family variants A1 and A2 of β -CN did not impact milk yield or composition. The κ -CN genotype AA resulted in lower ($P < 0.05$) milk protein content ($3.36 \pm 0.01\%$) compared to AB ($3.41 \pm 0.01\%$) and BB ($3.44 \pm 0.02\%$). The β -LG genotype influenced fat content, where AA ($3.59 \pm 0.04\%$) had lower ($P < 0.05$) values compared to AB ($3.65 \pm 0.04\%$) and BB ($3.67 \pm 0.04\%$). In conclusion, breeding for A2 cows does not impair milk yield or composition. Funded by grant PID2019-110752RB-I00 and the Ministerio de Ciencia Innovación y Universidades and supported by the Frisian Federation of Catalonia (FEFRIC).

Lipid oxidation and physical quality characteristics of beef as affected by storage condition

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The study was funded under the National Recovery and Resilience Plan (NRRP), of Italian Ministry of University and Research funded by the NextGenerationEU. The trial was aimed at studying three different preservation systems on the physical and nutritional characteristics of beef (sirloin cut). We compared two mixtures of modified atmosphere, MAP1, 80% O₂ – 20% CO₂, and MAP2, 60% O₂ – 20% CO₂ – 20% N₂, and vacuum conservation (skin, S). After packaging, beef was stored at 4 °C and sampled at three different periods; 7, 10, and 15 days. Samples were analyzed for pH, color, content of myoglobin, oxymyoglobin and metmyoglobin, water holding capacity (WHC), tenderness (shear force), lipolysis extent, profile of the volatiles organic compounds (VOCs) by SPME GC-MS technique, content of Malonaldehyde (MDA) by TBARS test, and the cholesterol oxidation products (COPs) by means of GC-FID. Results showed that storage systems and storage times significantly affected either the lipid's oxidation and some physical features of beef. TBARS test showed that the skin method was the best storing system in limiting fatty acid oxidation (0.16 vs 0.61 and 0.64 mg MDA/kg beef in S, MAP1 and MAP2, respectively). The brightness of the sirloin samples was lower for S than for MAP1 and MAP2. Regarding the storage time, lipid oxidation mainly occurred during the first 10 days of storage. MDA doubled from 7 days (0.33 mg/kg) to 10 days (0.66 mg/kg), when it reached a plateau. This trend was similar for color parameter (chrome and hue angle). In conclusion, the skin packaging was effective for limiting the lipid oxidation but was slightly worse than MAP in terms of color protection.

Session 17

Poster 14

Potential use of a pocket near-infrared spectroscopy device to directly discriminate local chicken meat

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The application of portable near-infrared (NIR) spectroscopy devices for analysing animal-derived products has recently increased. However, its efficiency within the poultry industry remains uncertain. This study aimed to evaluate the feasibility of a pocket-size NIR device to discriminate poultry meat. A total of 105 birds from 5 chicken breeds were enrolled in the study (Robusta Maculata; Bionda Piemontese (BP), cockerel and capons; Bianca di Saluzzo; Millefiori Piemontese; Eureka). Spectra were acquired directly on fresh thigh and breast muscles using a pocket-size NIR device working in reflectance from 740 to 1070 nm every 1 nm. Principal component analysis (PCA) and partial least square regression discriminant analysis (PLS-DA) were performed on the spectra using “stats” and “caret” packages of R software. Spectra pre-treatment were applied with “waves” package. In PLS-DA model, the dataset was split into a calibration (70%) and a validation (30%) set. Results revealed that PCA and PLS-DA models were able to distinguish chicken parts with a high accuracy (>99%). Despite the models were not able to discriminate among breeds, the PLS-DA correctly separated BP cockerel from capons (>97% accuracy). In conclusion, pocket-size NIR can be used to rapidly detect chicken parts as well as meat from entire and castrated animals.

Feasibility of near-infrared spectroscopy to predict dry matter digestibility in broilers

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Improving feed digestibility efficiency is critical to minimize nutrient losses and reduce environmental impact but its determination by wet chemistry limits their applicability. This study assessed the potential of near-infrared spectroscopy (NIRS) to predict Yb, Ti and dry matter digestibility (DMD) in broilers calculated from Yb, Ti, and polyethylene glycol (PEG). A total of 192 fecal samples were collected from 576 male Ross 308 strain chicks supplemented with TiO₂ (2 g/kg), Yb₂O₃ (50 mg/kg), and PEG (5 g/kg). Reference values for Yb and Ti were obtained through a coupled plasma optical emission spectrometer. PEG content was estimated through an ad-hoc NIRS prediction model. Samples' spectral information was collected every 2 nm from 1100 to 2500 nm. Reference data were matched with the spectral information to develop modified partial-least square prediction models with an external validation. The obtained coefficients of determination in external validation and the ratio of performance to deviation (RPD) were 0.67 and 2.00 for Yb, respectively, and 0.73 and 2.28 for Ti, respectively. The prediction models for DMD_{Yb} were highly unreliable (RPD = 1.93), whereas DMD_{Ti} model could be used for rough screening purposes (RPD = 2.41). However, DMD based on PEG-predicted content underlined the limitations of the calibration model developed with feces obtained during the adaptation period of the animals, where they received a different diet than the one during the experimental period. In conclusion, NIRS could be adequate as an initial screening tool for Yb and Ti content determination, and DMD based on Ti.

Session 17

Poster 16

Health status of Skopelos goats and its impact on milk yield

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The objective was to assess health status of Skopelos goats and its impact on milk yield. A total of 286 purebred adult goats were used. Their genetic background was the same but were raised in two different farms under different systems (intensive and extensive). Goats were randomly selected and prospectively monitored for two consecutive lactations. In both farms, daily milk yield (DMY) of individual goats was recorded every 50 days. Each goat was also subjected to detailed clinical examination for animal-based health indicators. A mixed linear regression model (SPSS v.26) was used to estimate fixed effects of age, farm, stage of lactation, year of sampling, body condition score (BCS), and occurrence of specific health issues on DMY; random effect of individual animal was considered. The most prevalent health issues in the intensive system were: i) udder asymmetry (16-52%), ii) body abscesses (8-36%), and iii) swollen mammary lymph nodes (1-34%). In the extensive system were: i) anaemia (5-50%), ii) udder asymmetry (11-48%), and iii) poor hair coat quality (7-47%). Across systems, lameness, contagious ecthyma, udder fibrosis, and udder asymmetry were associated with a significant decrease in DMY by 0.25 kg (18.2%, $p < 0.05$), 0.19 kg (14.1%, $p < 0.05$), 0.13 kg (9.5%, $p < 0.001$), and 0.09 kg (6.6%, $p < 0.001$), respectively. Age, farm, and stage of lactation were consistently associated with DMY across all cases ($p < 0.001$). In conclusion, udder health problems prevailed in both farms with negative consequences in milk production. Hence, results underpin the importance of regular monitoring udder health status and efficient management. This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No101000216.

Addressing the challenges of the European dairy beef production system

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The EU dairy beef production system is based on raising calves born in dairy herds, either non-replacement heifers or males, that are fattened for meat. The dairy beef production system is comprised of up to 11 million calves per year and potentially contributes to at least 40% of the beef meat produced, with expectations for future growth. The system is mainly categorised as 1), unweaned calves at slaughter (white veal), and 2), fattening of calves after the weaning period as rosé veal, young bulls, steers etc. These calves have been considered a by-products ('surplus calves') at the dairy farms, and there are critical emergent drivers and trends which indicate that there is a need for an autonomous dairy beef specific discussion space for facilitating its transition to a more responsible and sustainable production. It is evident that more dialogue is required concerning different aspects of the current production systems. The main challenges are environmental impact, calf genetics (pure Holstein or crossbred calves), improving the beef traits of calves from the dairy herd, inappropriate feeding at early ages, suboptimal transportation (of unweaned calves; from farm to slaughterhouse or to third countries for slaughter), and overuse of antimicrobials masking deficient management and nutritional practices that otherwise would result in unacceptable levels of morbidity and mortality. Finally, novel farming approaches and models will be required to meet present and future demands. Models will seek to address different developments (business models) and constraints and will integrate all parts of the value chain, including pathways for the better integration of females in dairy beef farming.

Session 18

Theatre 2

Longitudinal retrospective study of intake recovery and growth in unweaned dairy beef calves subjected to fasting, transport, and entire commercialization at farm arrival

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Data from 387 unweaned male dairy beef calves enrolled in 6 studies from 2020 to 2023 were used to evaluate the impact of fasting, transport, and overall commercialization (marketing and transport) on intake recovery and growth (ADG) at farm arrival. Calves were distributed in 4 treatments according to the different managements applied in the previous studies: calves from a sole dairy farm that were not fasted neither transported (CTR), calves from a sole dairy farm that were fasted between 9 and 19 h but were not transported (FAS), calves from a sole dairy farm that were transported and fasted for 19 h (TRA), and multi-origin calves commercially marketed and transported for 9 h (COM). At arrival at the rearing farm (day 0), calves were allocated individually, and body weight (BW) was recorded. Calves were fed milk replacer twice daily, and they had ad libitum access to concentrate and water. Concentrate (CI) and milk replacer (MRI) intake were recorded daily. Finally, BW was recorded on day 7. Data were analyzed with mixed models. An interaction ($P < 0.01$) between treatment and time was observed for CI. On day 0 CTR, FAS and TRA had similar CI, while COM had the lowest CI. On d 1, all treatments decreased CI except CTR. On day 7, while FAS and TRA recovered CI to similar levels than CTR calves, COM calves had 50% less CI compared with the other groups. The COM calves had the lowest ($P < 0.01$) ADG compared with CTR, FAS and TRA calves. In conclusion, the entire commercialization process of unweaned dairy beef calves impairs intake recovery and growth at farm arrival.

Total protein and hemoglobin levels of veal calves at arrival are associated with antibiotic use and mortality
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Adequate supply of colostrum, vitamins and trace elements at the dairy farm is crucial to enhance calf resilience at the fattening farm. Total protein (TP) is a good indicator of colostrum intake in calves beyond 14 days old. Blood hemoglobin (Hb) is a marker of iron supply coming from a premix which also supplies vitamins and trace elements having a role in supporting immunity. The objective of this prospective cohort study was to examine the correlation of TP and Hb level of calves with antibiotic use and mortality. The dataset contained weight (W), TP and Hb level at arrival and individual antibiotic use and mortality during fattening of 10923 calves arriving from January 2021 and April 2023. TP (n=2856) and Hb (n=10923) was determined in blood samples by spectrophotometry within one week after arrival at the farm. Data analysis showed positive correlations between TP, Hb and W (Pearson's R=0.18, P<0.05). TP negatively associated to mortality ($X^2=45.1$, df=8, P<0.001) and individual antibiotic treatment rate ($X^2=63.4$, df=8, P<0.001). Antibiotic treatment rate was reduced when TP>70 g/L and increased when TP <58 g/L. Mortality was significantly higher when TP<52 g/L. Hb was negatively associated with antibiotic treatment rate ($X^2=15.5$, df=7, P=0.03) and mortality ($X^2=27.6$, df=7, P<0.001). More antibiotic use was seen when Hb<5.5 mM and less mortality was observed when Hb>6.5 mM. Calves' weight, TP and Hb at arrival are positively correlated with one other and each individual parameter is negatively correlated with antibiotic use and mortality in the fattening period. This shows potential to use veal calf integration data to optimize colostrum and feeding management for surplus calves on dairy farms which has a positive effect on health and welfare later in calves' live.

A proteomics-machine learning approach to unravel the impact of protozoan infection on calves' intestinal morphology and development

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This study aimed to investigate the effect of *Cryptosporidium parvum* (*C. parvum*) infection on calves' intestinal mucosa at the molecular level using a proteomics approach combined with machine learning (ML) analysis. Newborn male Holstein-Friesian calves were orally infected (n = 5) or not (control group, n = 5) with *C. parvum* oocytes on day 2 of life. Total proteins were extracted from jejunum samples of slaughtered calves on day 8 post-infection and subjected to label-free quantitative proteomics. Differentially abundant proteins (DAP) between the infected and control groups were identified using random forest supervised ML algorithm in R and used for gene ontology analysis in STRING. Among the top 30 identified DAP, there was a decrease in abundance of villin 1 (VIL1), plastin 1 (PLS1), ezrin (EZR), catenin beta-1 (CTNBN1), usher 1C (USH1C), and cluster of differentiation 36 (CD36). Infection-reduced DAPs are located at the villus brush border and annotated to biological pathways including regulation of microvillus length, brush border assembly, and intestinal absorption of carbohydrates and fatty acids. This study provides a deeper understanding of a hierarchy in the molecular interactions that disrupt villus structure and absorption capacity which may interfere with calves' intestinal development in response to *C. parvum* infection.

Portable nanosensor for rapid detection and characterization of botulinum neurotoxins in cattle

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Clostridium botulinum toxins (BoNTs) are the primary cause of botulism, exhibiting the highest potency among bacterial toxins and posing a significant threat to both humans and animals. These toxins effectively block the release of acetylcholine, resulting in paralysis and, ultimately, death. However, the current method of using the mouse lethality bioassay as a gold standard test for diagnosis has certain limitations. In this study, we have successfully developed a reflective-based biosensor capable of detecting serotype B (BoNT-B) and serotype C (BoNT-C) toxins. This biosensor employs a competitive immunoassay approach combined with the evaluation of the toxins' endopeptidase activity. To determine the proteolytic activity of BoNT B and C, we utilized synaptosomal-associated protein (SNAP-25B) linked to a vesicle-associated membrane protein (VAMP-2) as the BoNT peptide substrate. The thermal oxide layer (SiO₂) surface was modified with specific toxins to enable the selective, sensitive, and real-time detection of target analytes. The biochemical reactions occurring on the oxide layer or the detachment of peptide fragments from the oxide layer amplified the reflectivity signals of each interferometer. The biosensor developed in this study demonstrates a low-level limit of detection of 0.43 pg mL⁻¹ for BoNT-B and 1.24 pg mL⁻¹ for BoNT-C. Further, the developed method was validated to confirm the specificity and selectivity of the designed platform against the respective toxins. Additionally, the capability of the developed probe was verified using extracted toxins, allowing for the accurate quantification of their concentration in the sample.

Session 18

Theatre 6

Effects of milk supplementation with different levels of soy protein isolate on growth performance and health indicators of the Holstein calves

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Protein is the most expensive and important nutrient to support efficient growth in young calves. In most cases, newborn calves are offered milk at around 10% of their BW and free access to a concentrate mix containing 18-22% crude protein (CP). Calculations showed that young calves suffered from a metabolizable protein (MP) deficiency of about 20-40 g/day, but even a higher CP content cannot meet the MP requirement, mainly due to the low intake of starter feed in the first days of life. We hypothesized that the addition of highly digestible protein sources in milk may compensate for MP deficiency. This study investigates the effects of supplementing milk with soy protein isolate (SPI) on growth performance, health, and inflammatory markers in dairy calves. A total of 36 Holstein calves were randomly allocated to the experimental groups (0, 20 and 40 g/d SPI). Supplementation lasted from 3 to 21 days, but measurements continued until weaning. During the supplementation period, calves fed SPI did not reduce their milk or starter intake, but DM and CP intake was increased. Protein supplementation early in life affected calf performance, and total DMI and ADG were improved by SPI. Calves fed 40 g SPI per day achieved the highest weaning weight (83.7 kg), the highest height at withers (96.4 cm), the best feed efficiency (0.67) and the fewest days with diarrhea. The values of the inflammatory markers decreased, but the malondialdehyde value was not affected by SPI. In conclusion, the performance of pre-weaned calves can be improved by adding SPI to the milk.

Impact of breed and colostrum intake on serum IgG, growth, health, and blood lymphocyte profiles in preweaning dairy and crossbred bulls

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This study aimed to explore the effect of colostrum intake and breed on IgG kinetics, health, growth, and blood lymphocyte profiles in Holstein and Holstein-Angus crossbred bulls. Calves (n=40; 10/breed/trt) were assigned to 1 of 2 trt for the 0h and 12h meals: LOW (2.5g of IgG/kg BW) or HIGH colostrum (5g of IgG/kg BW). Calves were then fed milk replacer at 6% BW twice daily until weaning from d49 to 70. Water and starter were provided ad libitum. Blood samples were taken every 2h from 0 to 24h, at 36h, and 48h to assess serum IgG concentrations. Fecal and respiratory health was scored twice daily to estimate disease incidence. Blood was collected from a subset of calves (n= 20) on d14, 35, and 84 for lymphocyte isolation and staining with monoclonal antibodies against several $\gamma\delta$ T and B cell subsets and analyzed by flow cytometry. Calves were weighed weekly to calculate average daily gain (ADG). Data were analyzed in SAS 9.4. Serum IgG was increased ($P < 0.01$) in the HIGH calves and there was no impact ($P = 0.17$) of breed, yet crossbred LOW calves had increased ($P = 0.04$) serum IgG when compared to Holstein LOW calves. No breed or trt differences were detected ($P > 0.10$) for ADG, days with diarrhea or respiratory disease. The proportion of IgM+ B cells expressing CD21 and CD32 tended to be greater ($P = 0.08$ and 0.05 ; respectively) in the HIGH calves, whereas LOW calves tended to have a higher proportion ($P = 0.08$) of $\gamma\delta$ T cells expressing WC1.1. However, breed did not impact ($P > 0.10$) lymphocyte profiles in the blood. These results suggest that increasing colostrum intake increases serum IgG and impacts blood lymphocyte profiles; however, breed had no effect on serum IgG, ADG, health or lymphocyte profiles.

Genetic parameters for calf resilience and milk feeding traits recorded by automated milk feeding machines and their relationship with bovine respiratory disease in North American Holstein calves

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The primary objectives of this study were to: 1) investigate the genetic background of milk feeding traits and bovine respiratory disease (BRD) in North American Holstein calves; and, 2) derive novel indicators of calf resilience based on variability in calf milk consumption. Data from 14,749 calves was collected daily over a period of 6 years and daily milk consumption and per-visit milk consumption, daily sum of drinking duration, drinking duration per-visit, daily number of rewarded visits, and total number of visits per day were recorded over a 60-d pre-weaning period. Additional traits were derived from these variables, including total consumption and duration variance, feeding interval, drinking speed, and pre-weaning stayability. The number of times a calf was treated for BRD was also evaluated. We compared repeatability and random regression models (RRM) to estimate genetic parameters for various traits. We observed low heritability estimates for feeding traits based on the repeatability models (e.g., daily milk consumption = 0.09; daily number of rewarded visits = 0.020), which improved with RRM (e.g., daily milk consumption = 0.46 [0.050, 0.680]; daily number of rewarded visits = 0.120 [0.030, 0.450]). Moderate negative genetic correlations between BRD treatment and feeding traits suggest potential for genetic improvement in disease resilience. We modelled and evaluated deviations in observed and predicted daily milk consumption trajectories as indicators of resilience to environmental perturbations. Parameters such as amplitude, perturbation time, recovery time, and deviation velocities were calculated to quantify the degree and duration of perturbations. We observed heritability estimates ranging from 0.16 to 0.52. This study provides key insights of the genetic background of early in-life traits in dairy cattle, which can be used for selecting animals with improved resilience, health outcomes, and performance.

Prevalence and distribution of foot lesions in French slaughter dairy and beef young bulls housed in indoor feedlot

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Nearly half of beef calves and 14% of dairy calves in France are reared indoors as feedlot cattle, where lameness is increasingly reported, especially towards the end of the fattening period. The objective of this study is to assess the prevalence and distribution of foot lesions at the end of the finishing period in French dairy and beef young bulls. A cross-sectional study was conducted in three regions with the highest proportion of feedlot cattle slaughtered in spring and autumn 2024. A total of 2102 animals were included in the study, representing various breeds: Charolais (56.8%), Limousin (11.7%), Blonde d'Aquitaine (6.9%), mixed breed (16.2%), Holstein Friesian (3%), and other breeds (5.4%). The 4 feet of each animal were trimmed and examined post-mortem by three trained professionals, with foot lesions categorized by type, severity, and location based on the ICAR Claw Health Atlas and national scoring methods. Descriptive analysis was conducted to estimate feet-, animal-, and within-batch-level prevalence, describe the severity and location of foot lesions, and estimate the distribution (e.g., breed, geographical area, etc.) of foot lesions. Preliminary results show that 34% of animals had at least one painful foot lesion, potentially indicating lameness. The most common painful lesions were stage M2 of digital dermatitis (affecting 17% of all animals) and level 2 of white line disease (present in 15% of animals). These findings might help raise awareness among stakeholders about the importance of lameness in young bulls, identify potential risk factors, and thus suggest possible control measures.

Session 18

Theatre 10

Updating preliminary study of multiphase diet for dairy beef cattle

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Crossbreeding between beef and dairy cattle has become a common practice in the dairy industry. However, there exists a lack of data and comprehensive calculations of the nutritional requirements for these crossbred animals, which hinders the implementation of effective precision nutrition techniques and limits the advancements in feed efficiency and sustainability within this specific production system. This study aimed to address this gap by investigating the nutritional requirements of crossbred dairy beef animals (i.e., F1 of dairy cows inseminated with beef breeds) and evaluating the application of multiphase diets as a precision nutrition technique. A preliminary analysis demonstrated the efficacy of multiphase diets in improving production costs, N excretion, and feed efficiency in cattle by changing dietary energy and protein concentration at three different points of the animals' growth. The current study expands the previous analysis by assessing a larger population of animals (n=16,445) in which we evaluated BW, ADG, and DMI with a particular focus on younger individuals (101-506 kg BW). To calculate the changing points, we first estimated the dietary density of protein required by dividing the metabolizable protein (MP) required by the observed DMI. Then, we performed regression analyses to adjust the data according to a reference diet (88 g MP/kg DM). Results showed a quadratic regression [$0.0248 - 0.631 \times BW + 0.00064 \times (BW^2)$], which fitted (the root of the mean square error of 34.68 kg) the MP required in the diet across the BW. The changing points were found at 353 and 436 kg BW, corresponding to 102 and 81 g MP/kg DMI, respectively. This result suggests that changing the diet at these points may improve the feed efficiency of dairy beef cattle.

Carbon footprint of Holstein bull calves fed two different total mixed rations from 4 to 12 months

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Despite the climate impact of rosé veal calf production is lower than most other beef types, it is still warranted that we find new ways of improving the climate impact of this production. The objective was to estimate the carbon footprint (CF) of rosé veal production when calves were fed two different feed rations. 64 Holstein male calves were fed and raised similarly from birth to 4 months, after which 32 calves were fed a corn-cob-silage-based TMR (YEL) and 32 calves were fed a grass-clover-based TMR (GRE) until slaughter at 12 months of age. YEL TMR also included barley, rape seed meal, and sugar beet pellets. GRE TMR also included barley, fava beans, and rape seed. Crude protein level was reduced twice during rearing. Feed intake (DMI and NEI) was similar for GRE and YEL. ADG from birth to slaughter and carcass weight were higher (268 vs 261 kg) for GRE vs YEL ($P < 0.03$). Tabulated values for CF of the various feedstuffs were used in the Life Cycle Assessment (LCA). Animals' methane production were measured in open-circuit respiration chambers at 8 months of age. The contribution of feedstuffs to CF of meat including soil C sequestration was 7% lower for GRE vs YEL. The contribution of manure was slightly lower for GRE vs YEL, but the contribution from enteric methane was 12% higher for GRE vs YEL. Total CF was 2861 vs 2858 kg CO₂e per calf produced. When CF was expressed per kg carcass, GRE was 3% lower (10.7 vs 11.0 kg CO₂e). Overall, this LCA confirms that rosé veal calf production has a low CF compared to other beef systems. The feeding effect shows that even a marked increase in enteric methane with one feeding can lead to a similar CF per kg meat if the contributions from feed, manure and C-sequestration counterbalance this.

Session 18

Theatre 12

Innovative sustainable organic beef production system where quality replaces quantity

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The GroBEat project proposes an innovative organic beef production system that offers a diversity of beef products (A, B, and C), is based on offspring from dairy cows, and which scores high on animal welfare, biodiversity and climate thus addressing specific criteria of major importance for the consumers. Furthermore, the high level of meat- and the sensory quality together with the storytelling of the system address that "less is more", which is supposed to facilitate healthier eating habits, while ensuring wellbeing, and providing the organic dairy sector with improved credibility for 'license to produce'. The GroBEat production system is based on steers and includes calf rearing by a foster cow for up to 6 months, use of low energy density roughage during winter, and utilizes extensive pasture for weaned steers. The system takes advantage of the variation in growth capacity between animals by slaughtering 1/3 of the best performing calves at 8 mo as high value rosé veal (A), another 1/3 of the steers at 16 mo as young beef (B), and the last 1/3 of the steers at 26 mo as mature beef (C). This gives three well characterized products compared with the current one product from intensive conventional rosé veal production and the traditional organic beef from steers slaughtered at 24-26 mo. The A steers were slaughtered at 347 kg LW, the B steers at 544 kg LW, and the C steers at 732 kg LW. The A steers spent 126 days, the B steers 267 days, and the C steers 367 days of their life on pasture. The A steers had the highest feed efficiency and the C steers the lowest. However, when considering the feed intake by the foster cow, feed efficiency of the A and B system did not differ. In the A system, 71% of the feed intake was assigned to the foster cow, 32% in the B system, and 20% in the C system. Finally, the system was evaluated both from a farmer perspective focusing on feed efficiency and economic return per animal and feeding day and from a society perspective focusing on carbon footprint and biodiversity per kg carcass using LCA.

Novel administration of meloxicam to surgically castrated bull calves via medicated lick blocks

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Livestock husbandry procedures such as surgical castration inflict long-term pain and inflammation, highlighting the animal welfare and production concerns. Pain relief products available to producers including topical anaesthetics (TA) and non-steroidal anti-inflammatory drugs (NSAIDs) are limited to peri or post-operative administration, failing to address procedural pain, and lasting only 24-72 hours. Fourty Bos taurus calves were randomly allocated to treatment groups: meloxicam lick block (ML), subcutaneous meloxicam (M), no meloxicam (NC), or sham castration (PC). ML calves had 19 hours of access to the medicated lick blocks prior to castration. M calves were administered 0.5mg/kg of meloxicam subcutaneously immediately prior to castration. Blood samples were collected from ML and M calves via jugular venepuncture for plasma meloxicam concentration (PMC) analysis. Weight, scrotal temperature and diameter, wound healing, and behaviour were measured. Meloxicam lick blocks reduced weight loss and increased locomotion and eating. On all days except the first, PMC was greatest in ML calves ($P<0.001$). The loss of weight was minimised in ML calves compared to M and NC calves ($P=0.044$). Eating increased in ML calves immediately after castration and was reduced when the medicated block was removed ($P<0.001$). Locomotion was greatest in ML calves ($P=0.018$). There was a trend for scrotal diameter to be less in ML calves compared to M ($P=0.069$). The administration of meloxicam via medicated lick blocks enabled pre-emptive and long-term analgesia. This project highlights potential for feed-based delivery of analgesia in numerous species.

Sustainability, Animal Welfare and Surplus Calves in the Australian Dairy Industry: Perspectives from the public – a focus group study

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Citizens are becoming increasingly disconnected from food production. Despite this, many people still hold strong values about how food is produced. The aim of this study was to attain an in-depth understanding of Australian public attitudes towards sustainability and animal welfare, as well as early life killing of surplus calves; an issue commonly identified as being out of step with public values. We conducted three, 3 hour focus group sessions, each with 7 Australians who varied in age, gender identity, income, and frequency of consumption of dairy products. Thematic analysis of the semi-structured discussions resulted in four key themes, each with underlying sub-themes: (1) sustainability, including concepts and behaviors seen as 'sustainable', consumer influence, and personal importance; (2) animal agriculture, including attitudes to farmers, and farming practices; (3) personal reflections, including emotions, ethical considerations, and knowledge and expectations; and (4) animal products, including consumption, alternative proteins, and health. Participants' attitudes towards sustainability and the consumption of animal products varied according to the different ethical lenses through which they viewed the topics, and alignment or otherwise with their personal values. Understanding and correcting where food animal production practices fail to align with the evolving core values of the public provides opportunities to preserve the social sustainability of animal agriculture into the future.

The use of gamma-glutamyl transferase as potential biomarker of colostrum consumption to evaluate the maternal antibody interference with subcutaneous and intranasal vaccines against BRD in unweaned dairy beef calves. An exploratory field study

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A total of 203 unweaned male calves were used to evaluate the use of gamma-glutamyl transferase (GGT) as biomarker of colostrum consumption on the maternal antibody interference with subcutaneous and intranasal vaccines against BRD in unweaned calves. At arrival to farm (day 0) calves were blood sampled to analyze GGT, IgG2, and IgM. After that, all calves received metaphylaxis against BRD using either subcutaneous (SC) or intranasal (IN) vaccines. Fourteen days after (day 14), another blood sample was collected to perform the same analysis. Data analysis was performed using Pearson's correlation with IgG2₁₄ IgM₁₄ and GTT₀ and mixed effects models with vaccine type (VT), GGT₀ categorized (GGT_{c0}) based on previous data (high-GGT ≥ 65 IU/L; low-GGT < 65 IU/L), and its interaction. The interaction between the GGT_{c0} and VT was not significant; however, low-GGT₀ calves tended to have greater (P = 0.11) IgM₁₄ concentration compared with high-GGT₀, and in IN vaccinated calves IgM₁₄ serum concentration did not increase (P < 0.01) compared with SC calves. In SC vaccinated calves, a negative correlation between GGT₀ and IgG2₁₄ (P < 0.001; r₂ = 7.8%; 95% CI -0.44, -0.1), and IgM₁₄ (P = 0.03; r₂ = 3.2%; 95% CI -0.37, -0.02) was observed, however the coefficient of determination was very low. These results highlight the potential use of serum GGT at arrival as a biomarker of colostrum consumption to identify potential maternal immunity by vaccine interference and better design effective vaccination strategies.

Session 18

Poster 16

Evaluation of milk replacer acidification according to the cattle origin as a feeding strategy during pre-weaning period on diarrhea incidence and performance in dairy beef calves

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Acidification of milk is a practical way to reduce diarrhea incidence and improve performance. This study aimed to investigate the effects of milk replacer acidification (MRA) depending on cattle origin (market vs. local) on pre-weaning performance and incidence of diarrhea in dairy beef calves. A total of 48 male calves (47 ± 6.6 kg of body weight and 24 ± 8.3 days of age) were used and allocated to treatments according to cattle origin (market vs. local) and feeding of MRA (acid vs. non), and they were weaned at 35 days of age. MRA was prepared to a target pH of 5.2-5.4 using formic and lignosulfonic acids. Calves were fed 2 L of milk replacer twice daily (125 g/L as fed) and had ad libitum access to concentrate and water. Animal body weight and feed consumption were recorded weekly. Fecal consistency was assessed daily using a 4-point scale; diarrhea was defined as fecal score ≥ 3. A subset of 20 calves was systematically selected for fecal sampling at days 0, 14 and 35 of study. Fecal samples were analyzed for enterotoxigenic Escherichia coli F5, Cryptosporidium parvum, Clostridium perfringens, rotavirus, and coronavirus using Bio-X immuno-chromatography test strips. Calves coming from market origin tended (P = 0.07) to show a greater incidence of Clostridium infection on day 35. No differences were observed in fecal score and days of diarrhea. There was an interaction (P < 0.05) between MRA and cattle origin in feed conversion rate with the acidification being more effective in calves from market. Thus, the efficacy of MRA was subject to the cattle origin.

Effect of a phytogetic feed additive (POWERJET®) on calves' growth performance and intestinal permeability
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The use of phytogetic feed additives has increased in animal nutrition to reduce the use of antibiotics and to improve zootechnical performance thanks to the combination of their active ingredients. The aim of the present work was to evaluate the efficacy of POWERJET® (PJ), a mixture of two plant extracts and their bioactive compounds with well-known antioxidant and anti-inflammatory properties in the Pharmacopoeia, on performance and intestinal permeability in milk-fed dairy beef calves. A total of 48 Holstein male calves (53 ± 3.6 kg of body weight, and 25 ± 9.5 days of age) were enrolled in a 2 x 2 factorial design, with the inclusion of PJ in milk-replacer at a dose of 2,000 g/t and in concentrate at a dose of 500 g/t. Calves were housed individually in boxes with straw bedding. Animals were fed following a milk-replacer program for 28 days (2 L twice daily, 125 g/L as fed), and had ad libitum access to concentrate and water. The trial lasted 56 days with two periods of 28 days each (pre- and post-weaning). Milk replacer intake and health status were recorded daily and concentrate intake and body weight weekly. Fecal consistency was assessed daily using a 4-point scale; diarrhea was defined as fecal score ≥ 3 . Blood samples were collected on days 0, 14 and 35 of study to determine the serum level of occludin protein as an indicator of intestinal permeability. No differences were observed in fecal score. Concentrate supplementation with PJ in comparison with no PJ tended ($P = 0.08$) to reduce serum levels of occludin (98.1 vs. 113.7 ± 9.60 ng/mL), reflecting a better preservation of the integrity of the intestinal epithelium. Calves fed milk replacer with PJ tended ($P = 0.08$) to improve feed efficiency during pre-weaning period (0.39 vs. 0.31 ± 0.032 kg/kg). This improvement was numerically observed over the study with PJ in milk replacer and concentrate (0.44 vs. 0.40 ± 0.028 kg/kg; $P > 0.10$). Thus, PJ inclusion in milk replacer and concentrate is a promising approach to ameliorate performance in dairy beef calves.

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Poster 18

Use of foster cows to produce rosé veal of spring-born dairy calves in an organic setting

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Most non-replacement calves born by organic dairy cows are sold to conventional rosé veal fattening. Due to a requirement of milk-feeding until 3 months (mo) of age, grazing, and at least 60% roughage in the fattening ration, very few calves are raised and slaughtered as organic, and only a few traditional steers slaughtered at 2½ years of age are produced within the organic system. Thus, currently no rosé veal of organic origin is available. Our aim was to develop and test a novel organic beef production concept producing three types of beef; 8-mo rosé veal (reported in this abstract), medium red beef from 16 mo-steers, and red beef from 26 mo-steers. The hypothesis was that performance as well as meat quality would be as good as similar types of conventional beef. To produce organic rosé veal we utilized culled dairy foster cows to rear the calves. A total of 24 Holstein cows each received two spring-born ½-mo old Holstein bull calves. Following successful bonding indoor and castration at 1 mo, cow-calf-pairs were grazing a grass-clover sward for 4 mo, and housed in the autumn. The 8 best-performing cow-calf pairs continued, and the 16 calves had access to the same TMR as the foster cows, were weaned at 6½ mo, and were slaughtered at 8.2 mo (ORG). There were no health-related issues with the calves. ADG from birth to slaughter was 1.23 kg/d, and carcass weight was 180 kg. Carcass, meat and eating quality was compared with 10 conventional 9-mo rosé veal calves (CONV, 211 kg carcass). EUROP conformation (ORG 3.2 vs CONV 3.6) and fatness (2.6 vs 2.4) were similar. Meat quality (intramuscular fat and colour measures: L*, a*, and b*) was similar. However, ORG had more intense taste and was more tender than CONV (both $P < 0.05$) with no difference in juiciness and smell. The study shows that it is possible to produce a high-quality rosé veal product in an organic setting.

Can cakes rich in polyunsaturated fatty acids substitute palm oil and reduce methane emissions in dairy beef calves fed high-concentrate diets? In vitro screening

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The aim of this study was to evaluate the rumen in vitro CH₄ production of concentrates containing different protein sources (PS) when partially replacing palm oil by different cakes (CK) rich in polyunsaturated fatty acids (PUFA). Concentrates' PS were: T1 = 8% soybean meal; and T2 = 15% dried distillers' grain. Palm oil (CTR, 4% and 3.5% for T1 and T2, respectively) was partially substituted (1%) by canola cake (CO; 6.5%), sunflower cake (SU; 14.5%) or camelina cake (CA; 11.5%), thus 8 types of concentrates were tested. Samples were grinded at 1-mm and used in an in vitro digestibility assay. The rumen liquid obtained at a slaughterhouse was strained through a cheesecloth and placed into pre-warmed thermal flasks at 39°C. Two incubation sets were done in parallel and in triplicate: two sampling points lasted 24 and 48 hours to calculate the DM digestibility; a third sampling point lasted 12 days to monitor the CH₄ production. Data were analyzed using a mixed effects model. Net volume of CH₄ per degraded DM did not differ between PS and CK (P=0.14), neither among CK, PS, and time (P=0.17). However, a tendency (P=0.09) was observed for PS where T1 tended to produce lower net volume of CH₄ per degraded DM than T2 (10.9 vs. 11.6 ± 0.45 ml/g, respectively). Partial substitution of palm oil by different cakes rich in PUFA did not reduce rumen in vitro CH₄ emissions.

Session 18

Poster 20

Effect of substituting palm oil by sunflower oil in fattening dairy beef bulls fed high-concentrate diets on performance and enteric methane emissions

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The aim of this study was to evaluate the substitution of palm oil by sunflower oil in dairy beef bulls fed high-concentrate diets on performance and enteric CH₄ emissions. Ninety-four bulls (399 ± 9.3 kg BW, and 271 ± 4.0 days of age) were group-housed in 6 pens and fed concentrate and straw separately, both ad libitum. Concentrate offered to bulls differed in the fat source: 1) CT (n=46), 3.5% palm oil; and 2) SU (n=48), 3.5% sunflower oil. The study lasted 42 days. Individual feed intake was recorded daily. Bulls were weighed every 2 weeks. Enteric emissions were recorded daily with CH₄ and CO₂ sniffers based on an infrared technology installed in each concentrate feeder and pen. Total daily CH₄ production (g/day) was estimated based on Madsen et al. (2010). Data were analyzed using a mixed effects model. No differences were observed in average daily gain (1.83 vs. 1.85 ± 0.141 kg/day, P = 0.87; CT vs. SU, respectively) and concentrate intake (7.48 vs. 7.48 ± 0.420 kg DM/day, P = 1.00; CT vs. SU, respectively). A significant time by treatment interaction was observed in enteric gasses emissions data with no clear pattern. Overall, mean CH₄ concentration per visit (73.0 vs. 91.6 ± 19.73 ppm, CT vs. SU, respectively; P=0.40); mean CO₂ concentration per visit (1442 vs. 1990 ± 576 ppm, CT vs. SU, respectively; P=0.39); ratio of CH₄ to CO₂ (0.050 vs. 0.046 ± 0.0100 ppm, CT vs. SU, respectively; P=0.80), and daily CH₄ production (184 vs. 175 ± 22.2 g CH₄/day, CT vs. SU, respectively; P=0.72) did not differ among treatments. When palm oil was substituted by sunflower oil in fattening dairy beef bulls fed high-concentrate diets, animal performance was not impaired and only a numerical enteric CH₄ emission decrease was observed.

Can algae rich in polyunsaturated fatty acids reduce methane emissions when substituting palm oil in dairy beef calves fed high-concentrate diets? In vitro screening

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The aim of this study was to evaluate the rumen in vitro CH₄ production of concentrates containing different protein source (PS) when partially replacing palm oil by different algae (AG) rich in polyunsaturated fatty acids (PUFA). Concentrates' PS were: T1 = 8% soybean meal; and T2 = 15% dried distillers' grain. Palm oil (CTR; 4% and 3.5% for T1 and T2, respectively) was partially substituted (1%) by *Nannochlorepis* (NA; 6.7%), *Chlorella* (CH; 9%), or *Tetraselmis* (TET; 13%), thus 8 types of concentrates were tested. Samples were grinded at 1-mm and used in an in vitro digestibility assay. The rumen liquid was obtained at a slaughterhouse. Two incubation sets were done: two sampling points lasted 24 and 48 hours to calculate the DM digestibility; a third sampling point lasted 12 days to monitor the CH₄ production. Data were analyzed using a mixed effects model. An interaction ($P < 0.01$) between PS and AG was observed in net volume of CH₄ per g of degraded DM; while no differences were observed for T1 (6.69, 6.30, 6.23, 6.31 ± 0.640 ml/mg; CTR-T1, NA-T1, CH-T1, TET-T1, respectively); in T2, CH-T2 produced greater net volume of CH₄ per g of degraded DM than the other T2 treatments, and NA-T2 produced lower net volume of CH₄ per g of degraded DM than TET-T2 (5.51, 4.79, 7.60, 6.06 ± 0.640 ml/mg; CTR-T2, NA-T2, CH-T2, TET-T2, respectively). Partial substitution of palm oil by different algae rich in PUFA did not reduce rumen in vitro CH₄ emissions.

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Poster 22

Carcass traits of Lidia breed heifers: preliminary results

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The Lidia breed of cattle is an autochthonous breed of Spain which grows extensively associated with a particular agrosilvopastoral ecosystem known as 'dehesa'. In this breed meat production has always been of secondary importance and has therefore been little investigated. Due to the challenges facing livestock farming with local breeds, and specifically the Lidia livestock systems, this work aims to make a first approximation to the characterization of the carcass of females of this breed. A total of 200 heifer carcasses (24-48 months old) from 10 farms (20 animals per farm) were analyzed. Animals followed a standard production cycle of the Lidia breed. Measures were obtained 24 hours post-mortem, on the left half-carcass and a descriptive statistical analysis was applied using XLStat software. The variables analyzed were: carcass weight, EUROP conformation and fatness score, morphometry (carcass length and depth, leg length and girth), pH₂₄, colour in CIELab* space of the M. rectus abdominis and subcutaneous fat and tissue composition of the 6th rib. The carcass of Lidia heifers was characterized by a lower weight, conformation and fatness compared to other breeds. Morphometry was similar to other local breeds and pH₂₄ was optimal. The colour of muscle and subcutaneous fat presented was darker, reddish and yellowish compared to carcasses commonly found in slaughterhouses. Tissue composition revealed a high bone content and medium lean and fat portions. The pasture-based production system, the high age at slaughter and the special rusticity and idiosyncrasy of this breed reflect the results found.

Session 19

Theatre 1

The 11th Modelling Nutrient Digestion and Utilization in Farm Animals (MODNUT) Workshop amidst the majestic Swiss Alps

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From 8 to 11 September 2025 will be held at Engelberg (Obwald, Switzerland) in the heart of Swiss Alps, the 11th workshop on modelling Nutrient Digestion and Utilization in Farm Animals (MODNUT). The local committee from Agroscope (Swiss centre of excellence for agricultural research) and international committee invite you to join the worldwide community of animal nutrition modelers. MODNUT occurs since 1979, when it started in United Kingdom, and since was held in North and South America, Oceania and Europe. Next year we will bring together researchers and active participants in the field of animal nutrition modelling, to share their findings and interact in the peaceful and studious atmosphere of Swiss Alps. The program will feature keynote lectures and topical sessions from basic static to intricate dynamic modeling across different levels of organization (cellular to farm), using mechanistic or empirical approaches. The 11th MODNUT will be followed by a one-day satellite workshop (12 September), with a focus on modelling processes and methodology including initiation to modelling software and practical examples applied to animal nutrition. Abstracts of this edition will be compiled in an Animal – Science proceedings issue, with the opportunity for presenters to publish a peer-review paper in a special issue of Animal (the international journal of animal biosciences) at special rate. The scientific program will be complemented by social activities to enhance networking opportunities and discover Swiss landscape and culture. We look forward to seeing you next year in Engelberg!

Session 19

Theatre 2

From the biochemical pieces to the nutritional puzzle: using meta-reactions in teaching and research

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We now live in an era where metabolic data are increasingly available. These data can be analysed using novel techniques (e.g., artificial intelligence) but it does not alleviate scientists to use “human intelligence”. The objective of this study was to extract information of a database of biochemical reactions with a method and tool to make nutritional biochemistry more accessible to nutritionists. A system of more than 300 meta-reactions (i.e., the outcome reaction of a series of connected individual reactions) was constructed covering a wide range of metabolic pathways for macro- and micro nutrients. Meta-reactions were constructed by identifying metabolic nodes, which are inputs or outputs of a metabolic system or that serve as connection points between meta-reactions. Complete metabolic pathways can be constructed by combining and balancing meta-reactions using a simple Excel tool, which has been used in teaching (e.g., to illustrate how much ATP can be synthesized from glucose, either directly or indirectly via storage and mobilization or via transfer). For research, nutrient balance data of the mammary gland of a dairy cow were used to construct a plausible pathway of nutrient metabolism of the mammary gland. The results highlighted the importance of the synthesis of proline from arginine and the concomitant synthesis of urea by the mammary gland. It also raised the question whether the availability of metabolic pathways or glucose uptake would be the more limiting factor for the synthesis of NADPH required for fatty acid synthesis. The availability of an open database of biochemical reactions, the concept of meta-reactions, and the provision of a tool allow users to acquire a more comprehensive and integrated view of metabolism and may raise issues that may be difficult to identify otherwise.

Hybrid local and global sensitivity analysis: Evaluation of dairy cow response predicted through INRA 2018 feeding system according to feed characteristics

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Two sensitivity analysis (SA) approaches were applied to the INRA 2018 feeding system to identifying the contribution of feed characteristics to the variation of the predicted animal responses. The SA has been performed in ruminant fields mainly by regression-based approaches that assume monotonicity of models. However, because INRA 2018 feeding system has complex models including interaction between feed characteristics, monotonicity cannot be secured. Thus, we applied not only differential-based local SA but also a variance-based global SA to evaluate our system. Five feed characteristics identified to affect animal responses prediction were chosen as input variables: organic matter digestibility (OMd), gross energy (GE), crude protein, and effective degradability and true digestibility of nitrogen. For the simulations, six reference diets for 2nd parity lactating cows at week 14 of lactation have been formulated. Overall, the relative sensitivities to input variables were consistent between both approaches. However, the sensitivity was different for dry matter intake (DMI) and milk protein yield (MPY) between diets. With grass hay-based diets, DMI appeared less sensitive to marginal variations in inputs with local SA, but showed a large interaction among input variables in global SA, while MPY showed the opposite trend. Regardless of the diet, the interactions took in mainly OMD and GE, the input variables that have the greatest impact on the energy/protein ratio that regulates DMI and MPY in the model. This work showed the advantages of a hybrid approach with two SA methods for analyzing ruminant feeding systems.

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Theatre 4

Predictive modelling of dry matter intake in lactating dairy cows based on routinely available variables

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Predicting dry matter intake (DMI) is crucial for farm management. The study aimed to develop a model based on readily available variables on a dairy farm. The data set collected in barn, between November and March for respectively 2015 to 2021, included 6202 weekly averaged daily observations in 413 lactations from 273 Holstein cows. Data were analyzed using R Statistical Software (v4.1.2; R Core Team 2022). A correlation analysis was performed to identify the best predictive variables. We identified five animal-related and two diet-related variables as the most correlated to DMI. Afterwards, a model was developed using backward regression from a full model consisting 7 variables. After removing non-significant variables (days in gestation, metabolic body weight, and dietary net energy of lactation), the selected predictive variables (mean \pm SD) were the following: parity 2.5 ± 1.6 ; Week of lactation (WOL) 14.9 ± 10.9 ; energy corrected milk (ECM) 33.7 ± 7.6 kg/d and neutral detergent fiber (NDF) 354 ± 59 g/kg DM. The metrics used for the evaluation were the root mean squared prediction error (RMSPE), its decomposition, and a concordance correlation coefficients (CCC) analysis was performed. The resulting prediction equation was $DMI(kg/d) = 16.86 - (0.18 \times Parity) + (0.24 \times ECM(kg/d)) - (0.02 \times NDF(g/kg DM)) + (0.29 \times WOL) - (0.005 \times WOL_2)$ (RMSPE = 1.45 kg, 7.16% and CCC = 0.90). Where parity is equal to 1 for multiparous and 0 for primiparous. In conclusion, our study successfully developed a predictive model for dry matter intake (DMI) in lactating dairy cows using easily accessible variables in dairy farms.

Dynamic sensitivity analysis of a mathematical model describing the effect of the macroalgae *Asparagopsis taxiformis* on methane production in a rumen in vitro continuous system

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Mathematical models have been developed to better understand rumen fermentation and methane (CH₄) production. The influence of input parameters (IP) of these models have been often assessed by analysis static sensitivity analyses (SA). We hypothesized that dynamic SA can be useful to inform on optimal experiments aimed at quantifying the mechanisms that drive CH₄ production. We used as case study a dynamic model representing rumen fermentation under in vitro continuous conditions. It describes the effect of *Asparagopsis taxiformis* (AT), a potent CH₄ inhibitor. We computed the Shapley effects (Owen, 2014) over time of 15 IP of the model to quantify their contribution to CH₄ (mol/h) variation. Two AT treatments were compared: a low treatment (LT: 0.25% DM of AT) and a high treatment (HT: 0.50% DM of AT). The IP studied were hydrolysis rate parameters (k_{hyd}, h⁻¹) of 3 polymer components in the diet, kinetic rate parameters (k_m, mol substrate/(mol biomass h) and K_S, mol/L) of 3 soluble substrates in the rumen, and parameters describing AT effects on fermentation (p). The hydrogen utilizers microbial group with IP K_S,H₂ contributed to more than 50% of CH₄ variation over time for control (0% DM of AT) and LT. For HT, IP p₂, which models the direct effect of AT on methanogens, showed a high influence (> 30%) over 2 time periods, highlighting a shift of the pathways impacting CH₄ variation over time. Further work is ongoing to exploit dynamic SA for designing optimal strategies using AT for CH₄ mitigation.

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Theatre 6

Using risk analysis to compare the accuracy of enteric methane emissions models of dairy cattle

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Many predictive models of enteric methane (CH₄) emission of dairy cattle were developed to help design effective mitigation strategies. Studying the precision and accuracy (Acc) of these models can provide recommendations on which model to use in specific situations. The precision indicates the confidence of a model in its prediction. While, the Acc indicates how far a prediction is from the true observed value. The Bayesian inference is useful to study them, as a prediction is represented by a probability distribution. This allows to compute the probability of a model prediction to be below a CH₄ emission (g/d) threshold. We developed 7 Bayesian models with data from de Ondarza et al., (2023) using predictors from Niu et al., (2018). The risk analysis was applied on 9 diet scenarios, determined by three levels (low, median and high) of dry matter intake (DMI, kg/d), ether extract content (EE, %DM) and neutral detergent fiber content (NDF, %DM) in the diet. The probability of models to predict the true CH₄ value of each scenario was compared to assess model Acc. Models using only DMI as predictor were the best and the addition of NDF improved their Acc. While, the addition of EE impacted negatively their Acc, except for high EE scenario (6.1 %DM). Considering NDF digestibility with DMI led to the highest Acc for median DMI and low and median EE scenarios. While, it showed the lowest Acc for low DMI and high EE and NDF scenarios. Further work is needed to enlarge the diet scenarios to study a wider range of variability.

Byproducts in Ruminant Feeding: Exploring their Mitigating Effects on Enteric Methane Emissions

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The increase in the availability of by-products (BPs) emanating from the processing of food to meet the growing population in different countries of the world has necessitated its exploration as feed resources in ruminant feeding. As part of the mandate of the ERA-NET joint call on circularity “INTEGRITY, ID – 83” project to evaluate different BPs of several origins and their level of inclusion in ruminant feed to enhance nutrient circularity and sustainability in the context of climate change. The specific objectives of this study were (i) to categorize identified BPs based on their chemical composition and their potential enteric methane mitigating effects and (ii) to develop specific methane prediction models. A database was created through a literature review using web search online databases (Science Direct, Web of Science) for articles from 2000 to 2024 using several keywords (methane, in vivo, GHG, BPs). The BPs were categorized into clusters using clustering analysis (k-means) based on similarities in dietary chemical composition (Ether extract, EE%; Crude protein, CP%; Neutral detergent fibre, NDF%, Starch, STA%). The elbow method at the flattening point with the lowest within-cluster sum of squares (WCSS) was used to determine the optimal number of clusters. The database contained 33 publications and 33 trials including 24 BPs fed to ruminants (n = 61 records) at different levels of inclusion. Methane was measured using SF6 (n = 10), Greenfeed (n = 10), Respiratory chambers (n = 34), and indirect calorimetric system (n = 7). The BPs were categorized into four clusters namely byproducts with (i) high Starch (27.1%), mid EE (13.9%), low CP (13.8%), and NDF (17.9%) [n = 1, Rice bran] (ii) high EE (51.6 ± 5.7%), mid CP (18.0 ± 1.77%) and NDF (26.0 ± 15.5%), low STA (0.25 ± 0.035%) [n = 2, Algal meal, Whole cracked rapeseed meal] (iii) high CP (37.8 ± 7.5%), mid STA (6.53 ± 1.42%) and NDF (24.3 ± 10.1%), low EE (7.10% ± 5.5) [n = 9, Soybean meal, Canola meal, etc.] (iv) high NDF (46.5 ± 15.5%), mid CP (12.5 ± 4.55%) and EE (8.3 ± 5.02%), low STA (2.13 ± 2.63%) [n = 12, Coconut kernel, soybean hulls, etc.]. Further work is needed to develop an enteric methane model integrating the level of inclusion (% of the whole diet’s chemical composition) of BPs

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Hybrid Intelligent Mechanistic-Dynamic Models: Mathematical Animal Nutrition Models to Predict Enteric Methane Emissions of Grazing Beef Cattle

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Rapid advances in animal data collection have been occurring for beef cattle grazing systems using precision livestock technology (PLT). Extensive rangeland systems lacking the physical (electricity) and communication (wi-fi) infrastructure required to operate PLT now utilize portable pasture-based systems, such as real-time weighing and enteric emissions monitoring, precision supplementation, virtual fencing, and behavior tracking. Mobile systems capture enteric emissions in the U.S. cow-calf sector. However, capturing continuous real-time individual animal data is challenging and does not reflect sub-daily changes in emission rates for individual animals. Interpolated or aggregated values may also fail to capture the dynamics of grazing systems (forage quantity and quality), management (rotational, continuous), animal classes (calf, heifer, cow), phases (growing, pregnant), and differences in feed and gain efficiency levels. Modeling is required to estimate emission rates at a regional scale more accurately. We propose a Hybrid Intelligent Mechanistic Model (HIMM) approach that leverages current principles to dynamic precision system modeling (PSM). Artificial Intelligence (AI) was used for the integration of covariates that contribute to individual changes in CH₄ from ~130 yearling Angus steers grazing native rangeland and monitored using a suite of precision technologies (real-time weighing, tracking, enteric emissions monitoring) and climate data. The AI model was then connected to a dynamic rumen kinetics model to create a HIMM model that predicts individual hourly enteric emission rates with acceptable accuracy and precision. Utilizing HIMM approaches will play a critical role in balancing the power of big data and AI with the mechanistic-dynamic modeling approaches that maintain the fundamental biological feedback linkages within grazing systems.

Systemic modelling application in animal science: archetypes for teaching and research in animal nutrition
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Systems perspectives and system dynamics (SD) have been used in decision-making and teaching in many fields. Their use in dairy nutrition and management, remains limited and, for the most part, unheard of. This work aims to present essential feedback thinking structures and behaviors, collectively known as archetypes in SD, but specifically designed for dairy applications. The models of feedback thinking are graphically illustrated using causal loop and stock-and-flow diagrams to show the ability of SD to contribute to complexity understanding in the field. Qualitative modelling was devoted to showing the role of loops in nutritional systems and the “Loop Dominance” concepts to explain and understand the feedback roles in determining the behavior of the system variables over time. Quantitative modeling was devoted to showing well-known structures that exhibit characteristic exponential, goal-seeking, oscillating, overshooting, or combined behaviors. The models have been built as the “System Herd”, from the example of the “System Zoo” draft of 1993 and from the book “Limits to Growth” published in 1972, by Donella Meadows. Simple structures were developed referring to one stock model (with examples on microbial population, passage rate, and feed intake) and two stock models (with interactions among pasture – grass or intake – reserves, digestive system descriptions, ruminal dynamics). Small complex models were explicitly built with emphasis on the effects of delays and accumulations and emerging behavior from multiple system elements. Examples from Agent-Based Modeling showed how to model emerging behavior from groups of individuals. These models represent a primary archive of basic structure operating under expected behavior with potential relevant role in SD teaching and applications for the animal science field.

Session 19

Theatre 10

Enhancing Agricultural Sustainability: An Optimization Model for Crop Rotation and Diet Formulation in Dairy Farms under European Union Common Agricultural Policy (EU CAP)

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In pursuit of agricultural sustainability, the European Union Common Agricultural Policy advocates for effective crop rotations. However, crop planning complexities, particularly for dairy farmers, are pronounced due to intertwined diet formulation. Our model is applied to a case study of a North Italian dairy farm (300 hectares, 45 fields, 1007 animals) to assess the impacts of three environmental policy scenarios aimed at mitigating enteric methane emissions: 1) MULTI: a multi-objective optimization that balances between economic gains and methane emissions; 2) TAX: applying taxation on methane emissions; and 3) RED: setting explicit methane reduction targets. Results revealed intrinsic trade-offs between economic returns and methane emissions across all scenarios. Adjusting the economic–environmental weighting factor resulted in a decrease in net revenue from 3167 to 1623 €/cow/yr, alongside a reduction in methane emissions from 131 to 117 kg/cow/yr. Methane mitigation prompted dietary changes, leading to diets with higher dry matter (from 47% to 65%), crude protein (from 14% to 16%), starch (from 21% to 23%), and reduced neutral detergent fiber (from 42% to 32%). This study underscores the potential of concurrently optimizing crop rotation plans and nutritionally balanced animal diets. By integrating both economic and environmental considerations, it offers valuable insights for dairy farmers, policymakers, and the broader agricultural community.

Assessment of amino acid requirements by crossbreed and sex of growing pigs based on sequential measurements of body weight and feed intake

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Individual body weight (BW) and cumulated feed intake per pen (CFI) were measured every 3 weeks between 69 and 155 d of age from 196 offsprings (either entire males (EM) or gilts (G)) born from a batch of Large White x Landrace sows inseminated with boar semen from two commercial lines (Choice, P81 or P90). Pigs were divided into pens (7 pigs each) according to crossbreed, sex, and initial BW. They were fed ad libitum with iso net energy diets (9.75 MJ NE/kg), providing 1.00 g standardized ileal digestible lysine per MJ NE during the first 6 weeks, and 0.90 g/MJ thereafter. Based on average BW and CFI obtained at different ages, 4 (sex × crossbreed) growth curves and feed curves were obtained using the InraPorc® software calibration procedure. Between 30 and 116 kg BW, average daily feed intake (EM: 2.52 vs. G: 2.62 kg/d, P=0.001), body weight gain (1119 vs. 1063 g/d, P<0.001), and feed conversion ratio (FCR; 2.27 vs. 2.47, P<0.001) differed significantly between sexes but not between crossbreeds. The corresponding growth profiles were similar for P81-F and P90-F, but different for ME: P81-ME being later maturing pigs than P90-ME. Subsequently, the amino acid (AA) requirement of P81-ME could be met with the same Phase-1 diet as G (0.89 vs. 0.95 g/MJ for P90-ME), and with the same Phase-2 diet as P90-ME (0.77 vs. 0.70 g/MJ for G). The study illustrates the usefulness of a nutritional model for estimating AA requirements of growing pigs from on-farm data and shows that similar average growth performance can be associated with different changes in requirements as BW increases, which is crucial to consider when designing optimized feeding strategies to improve pig efficiency.

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Theatre 12

OCalM: A new dynamic model to simulate the fate of calcium and phosphorus in laying hens

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The daily formation of eggshell is associated with several digestive and metabolic mechanisms. In order to design adapted mineral feeding strategies, we developed a dynamic model simulating calcium (Ca) and phosphorus (P) fluxes in laying hens over their productive career. According to diet characteristics and feed intake, digestive fluxes are first simulated, considering two different forms (fine vs. coarse) of dietary Ca in crop and gizzard. In the intestine, Ca and P are subsequently absorbed to the plasma, with a large increase in Ca absorption during eggshell formation. At the metabolic level, fluxes to/from the bone depend on egg requirements. Thus, after oviposition, Ca can be stored in the bone (accretion), while bone Ca can be mobilized when requirements for eggshell are important. This phenomenon especially occurs at night when feed is not available, thus liberating P which is excreted in the urine. These fluxes are then simulated over the animal's career, taking into account the evolution of laying parameters over time (laying rate, oviposition time). Finally, a "flock" module was added by considering three animal "profiles" ("good", "average" and "poor" layers). Simulations for different levels of coarse Ca in the feed are proposed, stressing out the importance of supplying Ca in a form allowing a progressive absorption over time, especially at night. However, the simulations confirm the role of fine Ca in replenishing bone reserves, especially just after oviposition. This model represents a new tool for nutritionists to design mineral feeding strategies preserving bone health in laying hens with long productive careers and limiting P excretion.

Evaluation of a mechanistic model predicting phosphorus and calcium requirements for bone mineralization of growing broilers

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As a non-renewable resource essential for both plants and animals, phosphorus (P) dictates our capacity to feed the world. To parsimoniously use it for meat production, a better understanding of the fate of dietary P by growing broilers is necessary. We therefore developed a first model to estimate the metabolic fate of calcium (Ca) and P in broiler chickens. Following inversion modelling principles, this model was inverted to predict requirement. Phosphorus requirements is the sum of the maintenance requirement and the amount that should be absorbed to reach soft tissue ash and bone ash potential. Requirements are the amount of Ca and P that must be absorbed to maximize the growth of soft and bone tissues, so those in “digestible” form. Different steps of model evaluation were performed. First, a global sensitivity analysis was performed. Then, the model was compared with requirement of Ross 308 and those of Rostagno et al. (2017). Also, the accuracy of the predicted requirements has been assessed in two trials. The sensitivity analysis for digestible P and Ca requirement showed that protein maturity ratio represented about 20%, while percentage of Ca in body ash represented 75% of the total variation observed. This shows the high requirement of these mineral for bone tissue. The results showed that the current model predict P and Ca requirements that are lower than Ross 308 especially in finishing period (about 22% for P and 7% for Ca) and lower than Rostagno (about 16 and 32% difference for P and Ca). Using this modeling approach, requirements can be defined based on different criteria of interest that depend on production objectives (e.g., profitability, animal welfare, and environment). Validation trials are ongoing and will allow adjusting the model before it could be used by nutritionists to dynamically predict the concentrations of those minerals in the feed of broiler chickens.

Session 20

Theatre 1

Effects of dietary supplementation with cocrystals of thymol and carvacrol on quality, nutrient composition, and oxidative stability of broiler meat

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Consumer demand for high-quality meat has become a rising trend. This study investigated the potential application of cocrystals of thymol and carvacrol in broilers for high-quality meat production. Eight hundred 1-day-old chicks were assigned to four groups fed the diets supplemented with 0, 10, 15, and 20 mg/kg Crystal EO® (CEO) containing 25% cocrystals of thymol and carvacrol in a 42-d feeding trial. Results showed that compared with the control group, dietary CEO supplementation significantly decreased muscle fiber diameter ($P < 0.05$) and significantly increased muscle fiber density, glycogen content, L^*45 min value, and proportion of α -linolenic acid of breast muscle ($P < 0.05$); dietary 10 and 15 mg/kg CEO significantly decreased lactate content, MDA concentration, cooking loss, shear force, and thrombogenicity index ($P < 0.05$), and significantly increased the proportion of lauric acid of breast muscle ($P < 0.05$); dietary 15 and 20 mg/kg CEO significantly increased glucose content, total superoxide dismutase, and total antioxidant capacity levels of breast muscle ($P < 0.05$). Especially, citrate synthase activity, free radical scavenging capacity, pH24h and a^*45 min values, and cystine content of breast muscle were also significantly higher in 15 mg/kg CEO group than those in the control group ($P < 0.05$). Moreover, all the variations changed significantly demonstrated linear and/or quadratic effects as dietary CEO supplementation increased ($P < 0.05$). Collectively, dietary CEO supplementation improved meat quality and nutritional values and enhanced antioxidant capacity of broiler meat, with 15 mg/kg CEO having the greatest effect.

Replacing Vitamin E in Broiler Diets with Polyphenolic-Rich Greek Vinification By-Products

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Grape pomaces from Savatiano, a native Greek *Vitis vinifera* white variety were collected after the vinification process. Grape pomaces were dried naturally and then the polyphenolic extract (GPE) was obtained using a methanol-water solution in ultrasonic and then was encapsulated. Wine lees were also derived after the wine-making process of a Greek *Vitis vinifera* variety. The resulting solid precipitate of wine lees after processing was used as a feed additive rich in yeast cell walls (WLYW). One hundred sixty, 1-day-old, Ross 308 broilers were allocated to 4 experimental treatments of 4 floors of replicate pens each. In the T1 group, vitamin E was adjusted to 75% of requirements while 0.1% of GPE containing 25 mg/kg feed polyphenols and 0.2% WLYW were included. In the T2 group, vitamin E was adjusted to 50% of requirements while 0.2% of GPE containing 50 mg/kg feed polyphenols and 0.2% WLYW were included. In the T3 group, vitamin E was excluded from the feed (0% of requirements) while 0.4% of GPE containing 100 mg/kg feed polyphenols and 0.2% WLYW were included. Finally, a Control (CON) group including 100% vitamin E requirements (80, 65, and 55 IU/kg feed in the starter, growing, and finishing phases, respectively) was allocated. Body weight, feed intake, and feed conversion ratio were recorded on the first day of the experimental period and at the end of each growing phase. At the age of 35 days, 12 broilers per treatment were randomly selected and sacrificed. Oxidative status, antioxidant capacity, and antioxidant enzymatic activities were assessed in blood and tissues. Carcass traits and meat quality were also assessed. Immuno-oxidative-related gene expressions were also determined in the liver, spleen, and bursa of Fabricius.

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Theatre 3

Modulation of broiler gut microbiota by dietary particle size, exogenous phytase and calcium concentration

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Particle size (PS) and calcium (Ca) concentration of feed affect the pH in the gut, which can modify exogenous phytase (EP) effects. Our goal was to investigate how PS, EP and Ca concentration impact gut microbiota. Eight experimental diets were formulated; the factors were PS fine (PF) and coarse (PC), Ca (4.9 g/kg and 7.2 g/kg) and EP (0 and 1000 FTU/kg). A total of 560 Ross 308 broilers were distributed into 56 metabolic units at 7 days of age and randomly assigned to each diet. On days 22 and 23, the birds were slaughtered, and the gizzard, ileum, and ceca samples were collected and pooled on a pen basis. DNA extraction was followed by 16S rRNA gene target sequencing. PC decreased α -diversity in the gizzard and increased it in the ceca compared to PF. In the ceca, EP and higher Ca led to an increase in α -diversity. A core community of 13 amplicon sequence variants (ASV) was found in the gizzard, ileum and ceca, and it represented more than 80% of the relative abundance in the gizzard and ileum and 30% in the ceca. Six of these ASV had significantly higher abundance in the PC group compared to PF in the ileum and ceca. Positive correlations that remained unaltered by the studied factors were found, i.e. *Lactobacillus-Limosilactobacillus* in the gizzard and ileum, *Clostridia* UCG 014-*Clostridia* vadinBB60, and *Eisenbergiella-Erysipelatoclostridium* in the ileum and ceca ($p < 0.05$). The results emphasize the effects of PS on diversity and community interactions in the ceca. EP and Ca increased the diversity of the ceca.

Effects of *Saccharomyces Cerevisiae* hydrolysate on Growth performance and gut health of broilers

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S. cerevisiae hydrolysate (SCH) is a promising alternative to antibiotics, due to its content of nucleotides, B-vitamins, amino acids, and polysaccharides. However, it is not clear how SCH modulates growth performance and health in broilers. This study evaluated the effects of SCH in wheat-based diets on growth performance, immune function, and intestinal health in broiler chickens. A total of 300 one-day-old male broilers were randomly assigned to 2 dietary treatments (C, basal diet; T, basal diet + SCH) and enrolled in a 42 days trial. Growth performance and intestinal health parameters were determined. Data were analyzed using one-way ANOVA and means were separated using Duncan's multiple range test (SAS). SCH significantly increased BW, ADG, and ADFI (d 1-42, $P < 0.05$). Villus height and the ratio of villus height to crypt depth of jejunum and ileum at day 28 and 42 were improved by SCH diet ($P < 0.05$). SCH down-regulated the mRNA expression of IL-1 β , IL-6 and TLR-2, and up-regulated ZO-1 and Occludin ($P < 0.05$). 16S rRNA analysis revealed that SCH altered cecum microbiota, increasing alpha diversity and the abundance of Bacteroidetes, while reducing Firmicutes. Taken together, SCH in wheat diets can stimulate the growth of broiler chickens by regulating the intestinal immunity and barrier function, improving the intestinal morphology, and relieving the intestinal inflammation, which may be related to the enhancement of bacterial diversity and the changes of intestinal microbial composition.

Session 20

Theatre 5

Carnosine in broiler diet: A promising nutritional strategy to reduce the prevalence of breast muscle myopathies

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The aim of this study was to assess the impact of dietary carnosine supplementation on growth performance, meat yield, quality and the occurrence of breast muscles myopathies. A total of 1,080 one-day-old male Ross 308 chicks were divided into two treatment groups with 12 replicates of 45 birds per each: 1) a control group that received a standard diet (Ctr) and 2) an experimental group that received carnosine supplemented at 500 mg/kg (Carno). Growth performance was assessed at the end of each growth phase. After slaughter, carcass and meat quality traits were evaluated. Muscles samples were taken to measure oxidation parameters and the antioxidant capacity. Breast muscle myopathies were visually assessed on 250 breasts per group. Results showed that carnosine incorporation in broiler diet did not alter production traits. Similarly, carcass yield, yield of carcass cuts and technological quality traits did not differ between groups. Interestingly, the carnosine supplementation significantly reduced the prevalence of white striping ($P < 0.001$), wooden breast ($P < 0.001$) and the co-occurrence of these two myopathies ($P < 0.001$). Furthermore, carnosine supplementation increased ($P < 0.001$) the antioxidant potential while malonaldehyde ($P = 0.30$) and carbonyl ($P = 0.80$) concentrations remained similar in groups. In conclusion, carnosine supplementation in broiler diet could be a promising solution for reducing the incidence of breast muscle myopathies and consequently minimizing economic losses in the poultry industry.

The effect of a phytogetic on broiler performance and feed costs in normal and reduced energy diets

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Phytogenics, specific plant metabolites, have demonstrated positive effects on metabolism, gut inflammation, and animal performance. Animal feed costs are a significant challenge in profitable broiler production. It is hypothesised that phyto-genics can improve growth performance in commercial diets and sustain growth performance in a diet with lower energy levels to reduce overall feed expenses. The trial was executed by BonÀrea Agrupa, with 3,072 1-day-old broiler chickens (ROSS 308) housed in groups of 132 birds/pen with similar body weight (41 ± 0.64 g), 8 pens/treatment. Three different dietary treatments were applied, with T1 being the control diet, T2 being the control diet with 25 ppm Fytera Perform (FP), a micro-encapsulated blend of clove, cinnamon, and oregano (Selko B.V., Tilburg, The Netherlands), and T3 being a diet formulated for lower energy and costs with 25 ppm FP. Data were analysed using a mixed-effects model with repeated measures. The results reported are for the entire production period (36 days of study). The body weight of T2 (2232.83g) and T3 (2247.70g) were higher compared to T1 (2192.48g) (SEM 12.93g, $P=0.02$). The average daily gain was significantly ($P=0.02$) increased in T2 (60.89 ± 0.357 g) and T3 (61.30 ± 0.357 g) compared to the control (59.76 ± 0.357 g). Daily feed intake was highest in T3 (94.11 ± 0.492 g) compared to T1 (91.04 ± 0.492 g) and T2 (92.35 ± 0.492 g) and feed conversion ratio was significantly ($P<0.01$) higher in T3 (1.54 ± 0.004) compared to T2 (1.51 ± 0.004) and T1 (1.53 ± 0.004). The feed cost per g growth was lowest in T3 (0.555 ± 0.0015 €/g growth) followed by T2 (0.559 ± 0.0015 €/g growth) and T1 (0.560 ± 0.0015 €/g growth) ($P=0.07$). In conclusion, FP improved growth performance in broiler chickens, with the diluted low energy/costs diet + FP resulting in similar growth as the control diet but at a lower cost.

Session 20

Theatre 7

Effects of yeast nucleotides administration via drinking water on broiler chickens

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Nucleotides actively participate in numerous biological processes and can become conditionally essential in periods of rapid growth or illness. This study aimed to evaluate the effect of yeast nucleotides (YN) administration via drinking water on growth traits and intestinal health of broiler chickens. 480 one-day-old chickens were enrolled in a 42-day trial and allocated to three experimental groups of 8 replicates each, where YN were included in the drinking water at a rate of 90 (T1) or 180 (T2) g/ton of water. The control group (CTR) received no supplementation of YN. Growth traits were evaluated on d 0, 10, 21, and 42. At d21 and 42, three animals per replicate were sacrificed; jejunum samples were collected for morphology and gene expression evaluation, while ceca content was collected for microbiota analysis. Multiple means were compared by one-way ANOVA (GraphPad Prism); $P<0.05$ was considered statistically significant. YN improved body weight at d21 and 42 ($P<0.05$). Average daily gain was increased in the period 10-21d ($P<0.05$) and over the entire trial ($P<0.05$). At d21, both T1 and T2 showed an improved villus:crypt ratio than CTR ($P<0.01$); a similar trend was observed also for villus height in T2 ($P=0.07$). T2 showed decreased mucus thickness at d21 compared to CTR ($P=0.03$). Analysis of microbiota and gene expression of tight junction and immune-related genes are currently being performed, which could help to better elucidate the role of YN in supporting gut health and performance of broiler chickens. Acknowledgment: this study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

Vertical transmission of gut bacteria in chickens and the implications of growing chicks in the absence of adults

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The gut microbiota performs a number of functions for the host, including converting plant derived nutritional fibers into short chain fatty acids (SCFA) which the host utilizes as an energy source, protecting the host from gut pathogens, and affecting the maturation and functioning of host systems, such as the intestinal, immune and neural systems. Thus, early establishment of the gut microbiota is required for optimal functioning of the microbiota. In nature a main mechanism for the transfer of gut bacteria from one generation to the next is the direct interaction between parents and offspring. However, in commercial settings chicks never meet an adult. To characterize vertical transmission, in conditions in which chicks are grown in the absence of adults, and to determine if the egg was a vehicle for vertical transmission, we compared the fecal microbiome of broiler breeder hens and their progeny till the age of two weeks. We found that most bacterial strains found in the hens did not appear at all in chicks up to two weeks of age. Furthermore, most strains common with the hens which did appear in chicks had a low incidence among the chicks. Thus, the gut microbiota of hens does not efficiently transmit in commercial settings. That been said, a few bacterial strains common with the hens were good colonizers of chicks. To conclude, vertical transmission in commercial poultry grown separately from hens likely exists but is not efficient, possibly resulting in impairment of microbiota function. To understand if microbiota functions were affected, we exposed newly hatched chicks to the gut content of adults which contains many bacterial strains. We found many adult bacteria colonized the chicks. We also found host gene expression in cecal tissue as well as in the bursa was different in treated chicks compared to control chicks. Furthermore, SCFA levels were much higher in treated chicks, implying treated chicks were better utilizing their feed. Thus, growth in the absence of adults has an impact on commercial chicks. Future work is needed to identify specific bacterial strains that positively impact chicks for development as probiotics.

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Theatre 9

Development of eco-friendly feeding approaches in poultry. Assessment of different dietary zinc sources

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Zinc (Zn) supplementation is a usual poultry feeding strategy since it can improve certain aspects of avian production. However, a Zn dietary excess can result in environmental damage, as the fraction that is not absorbed is excreted in the faeces. The purpose of this study is to determine the most suitable dietary zinc source by comparing the effects of Zn in the form of sulphate (Zn-S), glycine (Zn-G), and hydroxychloride (Zn-H) on quail performance, egg production, mineral excretion and mineralization of bone. A total of 75 female quails (10 weeks old) were randomly distributed into 3 groups (5 replications/5 females quails). Treatment groups were set up by adding Zn-S and Zn-H as the inorganic form and Zn-G as the organic form of zinc to the corn-soybean basal diet (34.14 mg/kg Zn) to obtain 50 mg/kg of Zn. The feeding experiment lasted for 12 weeks. Performance parameters and egg production were not impaired by the Zn source ($P > 0.05$). Most of the minerals assessed in the tibia were higher ($P < 0.01$) in quails fed with Zn-H compared to those received Zn-G and Zn-S which could be reflected in an improvement of bone health. Lower levels of mineral excretion were observed in Zn-G and Zn-H groups compared to Zn-S. It can be stated that Zn-H or Zn-G supplementation could be an interesting strategy to reduce mineral excretion without affecting performance compared to Zn-S. However, further studies are needed to clarify the differences between Zn-G and Zn-H supplementation on bone health.

The inclusion of microalga *Dunaliella salina* in the diet of laying hens: effects on production performance and egg quality

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Dunaliella salina is a microalga known for being a rich source of protein and carotenoids. This feedstuff has the potential to improve the productive performance and quality of table eggs. Additionally, *Dunaliella salina* can be produced locally with fewer resources. The study involved 32, 58 weeks old, H&N Brown Nick laying hens, monitored over a 7-week period. The hens were divided into four homogenous groups of 8 replicates with 4 levels of inclusion: 0, 1, 2.5 and 5% *Dunaliella salina*. The study assessed productive performances and egg physical characteristics of the eggs. Data were statistically analyzed using the One-Way ANOVA procedure of SAS with Tukey Test to identify the significant differences between means. The results indicated that the incorporation of *Dunaliella salina* did not significantly affect egg production and feed conversion ratio ($p > 0.05$). However, it was observed that the addition of 1% and 5% negatively impacted feed intake and egg weight, respectively ($p < 0.05$). The main physical quality parameters of the eggs, such a Haugh units, shape index, yolk index, egg surface area, shell index, shell thickness, and specific gravity were not affected by the addition of microalgae ($p > 0.05$), however, there was a negative impact on albumen pH ($p < 0.05$). The inclusion of *Dunaliella salina* in the diet led to an increase in both the redness (a^*) and yellowness (b^*) of egg yolks ($p < 0.05$). These findings indicate that the incorporation of *Dunaliella salina* in the diets of laying hens may affect some performance parameters, but not affect egg quality. Keywords: Egg quality, laying hens, microalgae, *Dunaliella salina*.

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Theatre 11

Unveiling the Immunomodulatory Potential of Probiotic *Bacillus* Strains in Chicken Peripheral Blood Mononuclear Cells

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Feeding probiotic *Bacillus subtilis* DSM 32315 (BS) and *B. velezensis* CECT 5940 (BV) enhances chicken health and performance, possibly through immune modulation. We investigated their effects on chicken peripheral blood mononuclear cells (PBMCs) via transcriptome profiling and cytokine array. Both treatments significantly altered gene expression, with BS affecting four immune pathways and BV affecting five. In both BS-treated and BV-treated PBMCs, the expression levels of T cell marker genes CD4, CD25, and CD28 were elevated ($p < 0.05$ for CD4, $p < 0.01$ for CD25, and $p < 0.05$ for CD28). Additionally, the expression of T cell-associated CC chemokine ligand 5 (CCL5) was increased in both BS and BV-treated PBMCs ($p < 0.05$), while the expression of pro-inflammatory IL16 decreased ($p < 0.01$). Furthermore, BS treatment resulted in an increased expression of anti-inflammatory cytokine gene IL10 ($p < 0.01$), whereas IL10 expression was not affected by BV treatment. The concentrations of cytokines in the culture supernatants remained unchanged in both treatments. Additionally, both BS and BV treatments led to an increase in the expression of avian β -defensin 1 (AvBD1) compared to the control ($p < 0.01$ for BS and $p < 0.05$ for BV). These findings suggest that both BS and BV treatments modulate immune responses in PBMCs, potentially enhancing immune function and host defence mechanisms.

Chicken immune cell assay to evaluate the immune-modulating effect of grape by-products

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Agro-industrial by-products offer sustainable and cost-effective alternatives to traditional feedstuffs. Grape by-products (GP) come from winery and are known to be rich in phenolic compounds. We evaluated the immunomodulatory effect of GP on chicken immune cells by isolating peripheral blood mononuclear cells (PBMC) from chicken blood and exposing them to various GP forms. Original GP (oGP) was digested *in vitro* using the INFOGEST protocol to obtain digested GP (ivdGP). Phenolic compounds were extracted from oGP using a water:ethanol solution, yielding extracted GP (extGP). We measured total phenolic content in oGP and extGP with a colorimetric Folin assay. Chicken PBMC were treated for 24 h with oGP, ivdGP, and extGP, and immunophenotyping was applied using flow cytometry. The extGP treatment increased the proportion of dead cells but significantly enhanced CD4+CD25+ and CD8+CD25+ T cells activation, suggesting that extGP induced T cell activation without promoting proliferation. No significant changes were observed in CD4 and CD8 T cell activation with other treatments, nor did any treatment significantly affect B cells. To conclude, only extGP enhanced CD4 and CD8 T cell activation among the tested GP forms, highlighting its potential immunomodulatory role in chicken PBMC immune response. This suggests that specific GP forms can influence the immune system, providing insights for their use for poultry nutrition and health management.

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Theatre 13

Investigating the impact of Sodium Butyrate on gene expression profiles in chicken intestinal organoids

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The advent of animal organoid technology presents significant research avenues in zootechnical and veterinary sciences. Of particular interest is the utilization of intestinal organoids, known as enteroids, in investigating host-microbiome interactions. Butyrate, a metabolite produced by probiotic bacteria and one of the short-chain fatty acids, holds potential to influence intestinal tissue integrity and its secretory functionality. The objective of this experiment was to culture *ex vivo* chicken intestinal organoids, stimulate them with sodium butyrate, and evaluate changes in transcript levels of genes associated with intestinal barrier function and immune response. Jejunal crypts were isolated from ROSS 308 chicken embryos (n=12) at day 18 of embryonic development. The crypts, containing intestinal stem cells, were purified, suspended in culture medium, and embedded in Matrigel. On the fifth day of *in vitro* culture, sodium butyrate was added to the culture medium at two concentrations: control-none, SB1–0.5mM, and SB2–1mM. After 24 hours, RNA was extracted from the organoids, and relative gene expression analysis was performed. Genes examined included interleukins (IL-6, IL-8, IL-17) and genes associated with intestinal barrier function (Cadherin, Tight Junction Associated Protein, Claudin 2, Mucin 2, Villin, Occludin). Kruskal-Wallis test was conducted to analyze the data, and intergroup comparisons were made using the Two-stage linear step-up procedure of Benjamini, Krieger, and Yekutieli test. The SB2 group exhibited a significant increase in pro-inflammatory cytokines IL-6 and IL-17, alongside a decrease in IL-8 levels with both doses. Additionally, two tight junction-related genes, OCLN and CLDN 2, were significantly upregulated with the higher dose of sodium butyrate. These preliminary findings indicate the effective stimulation of chicken intestinal organoids with sodium butyrate, as evidenced by alterations in mRNA levels of genes involved in inflammatory responses and tight junctions. Financially supported by the Minister of Science under the program “Regional Initiative of Excellence” (RID/SP/0017/2024/01) and National Science Centre (Poland) by grant 2023/07/X/NZ9/00241

Supplemental effect of *Aspergillus oryzae* phytase on growth performance, nutrient digestibility, and health parameters in Ross 308 broilers

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In feedstuffs of plant origin, phosphorus is present in both organic and inorganic forms. Most of the organic phosphorus in plant ingredients is bound to complex structures called phytate (Myo-inositol hexakisphosphate), the mixed salt of phytate. Though earlier studies showed a beneficial effect of adding exogenous phytases to swine and poultry, to date very limited studies have been presented on adding novel bacterial 6-phytase (Ronozyme HiPhos) expressed in *Aspergillus oryzae* to broilers. Therefore, we conduct this study to examine the effect of phytase on the overall performance of the broiler. Ross 308 (n=1512) chicks (48.05 ± 0.52 g) were allocated to four treatment groups with 21 replicates of 18 birds/pen. The test treatments included a basal diet (CON) and the CON diet enriched with 0, 500, 1000, and 1500 FTU/kg *A. oryzae* phytase. Experimental data were analyzed using the GLM procedure of SAS and the Linear, quadratic, and cubic test was used to determine the significance between treatment means (P<0.05). During starter (days 1-9), grower (9-21), and overall period (days 0-32) broilers fed a diet supplemented with 1500 FTU/kg of *A. oryzae* phytase demonstrated higher (P<0.05) body weight gain and feed intake compared to CON. However, there was no difference observed in nutrient digestibility, organ weight, footpad, and intestine lesion score. In conclusion, adding 1500 FTU/kg of *A. oryzae* phytase to a broiler diet would be beneficial to enhance their growth performance by releasing phytate-bound nutrients.

Session 20

Poster 15

Growth performance, nutrient digestibility and intramuscular fatty acid composition of chickens fed Camelina Cake

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The objective of the study was to investigate the influence of different amounts of camelina cake in chicken feed on their growth performance, nutrient digestibility and meat fatty acid composition. The base part of the control group (C) diet consisted of corn and soybean meal, while in the diets of the experimental groups the soybean meal was replaced by 5% camelina cake in the CAM5 group and 10% in the CAM10 group. Study results showed that the growth performance indicators of CAM5, CAM10 groups chickens did not differ from the C group. Although the digestibility of dry matter (P ≤ 0.01), organic matter (P ≤ 0.01) and protein (P ≤ 0.05-P ≤ 0.01) was lower in the CAM5 and CAM10 groups, the inclusion of camelina cake positively impacted meat fatty acid composition. We found significantly higher levels of various beneficial fatty acids in CAM5 and CAM10 groups: the muscles of the chickens of experimental groups contained 2.49 – 3.80 (P ≤ 0.01) -linolenic, 1.7 – 2.67 eicosapentaenoic (P ≤ 0.01), 1.83 – 2.28 docosapentaenoic (P ≤ 0.01), 1.73 – 2.17 docosahexaenoic acids (P ≤ 0.05-P ≤ 0.01), 1.72 – 2.47 total LC n-3 PUFA (P ≤ 0.05-P ≤ 0.01) and 2.29 – 3.36 total n-3 PUFA (P ≤ 0.01) times more than in the muscles of the control group. Also, significantly better n-6/n-3 and linoleic /linolenic ratio, as well as thrombogenic index, were found in the both experimental groups (P ≤ 0.01). The optimal n-6/n-3 PUFA ratio was determined in CAM10 group and according to the European Commission Nutrition Claims, the meat of chickens grown in the CAM10 group can be classified as a source of Omega-3 fatty acids based on the ALA content.

Silvafeed® ATX efficacy in improving performance of broiler chickens

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This study aimed to determine the effect of dietary Silvafeed® ATX levels (SilvaTeam – Itália) on performance and carcass yield of broiler chickens. 1,280 day-old Ross AP91 male broilers were distributed in four treatments (40 chickens/pen; 8 pens/treatment): (Control) and three levels of the Silvafeed® ATX: 250, 500, 1,000 g/t. Corn/soybean treatments diets were fed a pre-starter 0-7d, starter 8-21d, grower 22-35d and finisher 36-42d. Feed intake (FI), body weight gain (BWG), feed conversion ratio (FCR) and production efficiency index (PEI) were measured on day 42. At 42 d two birds per pen were selected for slaughtering and to measure carcass yield and its parts. The statistical assumptions for normality were verified and data were submitted to analysis of variance and significant estimates were subjected to regression analysis. FI showed a decreasing linear curve, which decreased with increasing Silvafeed® ATX levels. BWG showed a maximum gain point at the level of 664 g/t. FCR equation showed a better point at the level of 763 g/t and the maximum point of efficiency production index was 514 g/t of Silvafeed® ATX. There was no significant difference for the diet addition of Silvafeed® ATX on carcass yield. Results indicated that addition of Silvafeed® ATX supplied in the diet from 1 to 42 days of age contributes to improve broiler's performance according production efficiency index.

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Poster 17

Influence of L-Glu and L-Gln supplementation in broilers subjected to an Eimeria challenge on performance
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Dietary glutamate (L-Glu) supplementation was shown to improve growth performance in broiler under sanitary challenge and glutamine (L-Gln) improved performance by maintaining an optimal antioxidant status and immune function. L-Glu and L-Gln being closely linked metabolically, a supplementation of both amino acids may have a synergistic effect and would modulate growth performance and resiliency of broiler under Eimeria infection. This trial aimed to study broiler chicken response to L-Gln and L-Glu supplementation during an Eimeria challenge. A total of 1800 male day-old Ross 708 chicks were allocated to six treatments (12 replicate pens per treatment): 1) negative control (NC), 2) positive control (PC): NC+anticoagulants, 3) NC + 5g/kg L-Gln (100Gln), 4) NC + 3.3g/kg L-Gln + 1.65g/kg L-Glu (66Gln+33Glu), 5) NC + 1.65g/kg L-Gln + 3.3g/kg L-Glu (33Gln+66Glu) and 6) NC + 5g/kg L-Glu (100Glu). At day 10, half of the pens were challenged with oral gavage of 1mL of an Eimeria culture while the others received equal volumes of 0.9% saline. Performance data (body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR)) were subjected to a one-way ANOVA with diet as fixed factor for the pre-challenge period and a two-way ANOVA with fixed effects of diet, challenge and their interaction using a mixed model with the pen as random effect. The interaction was removed from the model if not significant ($P>0.05$). During the pre-challenge period, birds receiving PC diet and all supplemented diets with Gln and Glu had improved BWG and FCR compared to birds fed with NC. During the challenged period, challenge had a strong and significant effect on performance with challenged birds having decreased BWG, FI and increased FCR compared to unchallenged birds. No interaction was found between dietary treatment and challenge. For the overall period (0-35d), the birds fed PC and 33Gln+66Glu had a significant better FCR compared to the birds receiving NC, the other Gln/Glu combinations being intermediate. In conclusion, results demonstrated that the combination 33Gln+66Glu performed as well as the PC and better than the NC, whatever the challenge.

Production results and carcass characteristics of Cherry Valley broiler ducks fed with halloysite

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The study aimed to assess the production results and carcass characteristics of broiler ducks fed with 1% halloysite added to the feed throughout the 42 days of rearing. Ducks (100 males and 100 females) were divided into 4 groups according to sex—control groups – CM and CF, and experimental groups – ZM and ZF. In the experimental groups, broiler ducks were fed with 1% halloysite. The birds' body weight (BW) and feed intake (FI) were controlled, and production efficiency indicators were calculated. Analyses of the chemical composition of feed were performed. On day 42, the birds were slaughtered. The carcasses with offal (heart, liver, gizzard) were plucked and gutted. After cooling the carcasses, dissection was performed. Each part of the carcass was weighed. The data were processed using Statistica statistical software. Mean values and standard error of the mean were calculated. Tukey's test with $P \leq 0.05$ verified the significance of differences. In groups where halloysite (HM, HF) was used, significantly lower birds' BW was observed throughout the entire rearing period ($P \leq 0.05$). Broiler ducks in the experimental groups were characterized by significantly higher FCR from the 1st to 28th day of rearing ($P=0.003$). The highest liver weight was found in the HM group. Lower pectoral muscle weight was demonstrated in the HM group compared to CF and HF ($P=0.009$). The use of 1% halloysite in feed had no beneficial effect on selected production results, possibly due to its ability to limit dietary protein availability. The research was carried out as part of project No. UMO-2021/43/D/NZ9/01756, financed by the National Science Center (Poland).

Session 20

Poster 19

Assessment of Natural Silicate-Based Minerals Effects on Compound Feed Pelletizing Processes

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The aim of this study was to assess the effects of the inclusion of several natural silicate-based minerals during poultry compound feed processing, and more specifically on pellet durability index (PDI, %) and specific energy consumption (kWh/t). The tested minerals included three grades of bentonite (Bent_A, Bent_B and Bent_C), two grades of diatomite (Diat_A and Diat_B), two grades of moler (Mol_A and Mol_B) and one grade of kaolin (Kao_A). A wheat based broiler feed (19.8 % CP, 40.0 % starch, 5.5 % fat, 12.2 MJ/kg ME) was pelletized without any mineral (CON) or with the incorporation of 1.0 % DM of the different minerals. The pelletization of the feed was carried out with a pilot scale press (Amandus Kahl, Germany) using a fixed flat die (4.0 mm diameter), with the conditioner temperature set at 60°C. The PDI was measured using the Eurotest device (Sabe, France), while the specific energy consumption was estimated based on the press power (kW) and pellet output (kg/h). The PDI was significantly ($P < 0.001$) affected by the inclusion of minerals, the highest PDI was registered with Diat_B when compared to CON (72.0, 72.8, 72.8, 74.6, 81.0, 75.1, 76.7, 75.9 and 72.5 % for Bent_A, Bent_B, Bent_C, Diat_A, Diat_B, Mol_A, Mol_B, Kao_A and CON, respectively). The specific energy consumption was also affected by the inclusion of the different minerals, with Diat_B showing the lowest consumption when compared to CON (13.7 and 15.2 kWh/t). Based on these results it can be concluded that the inclusion of natural silicate-based minerals in a high-fat content broiler feed during processing could improve pellet quality while having a positive effect on energy consumption during production. Nevertheless, further trials are recommended in order to assess the effect of mineral inclusion in upscaled industrial production.

Effect of dietary micelle quercetin on growth performance, egg production, and blood profile in Hy-line brown laying hens

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Quercetin (QR) is commonly known as oak essence and naturally occurring polyhydroxy flavonoid found in fruits and vegetables. It exhibits a positive impact on egg-laying performance. Previous studies pointed out that QR acts as a potential alternative to antibacterial drugs however the implementation of Micelle form of quercetin (MQ) in poultry diet is still limited. Therefore, this study aims to assess the impact of MQ on growth and egg production performance in Hy-Line brown layers. For 12 weeks, laying hens (n=288) were randomly assigned to one of three test treatments: basal diet (CON) and CON diet supplemented with 0%, 0.03%, and 0.06% MQ. Each treatment has 8 replicates of 12 hens. Growth performance, egg production, and blood profile data were analyzed using the GLM procedure of SAS, and the Linear and quadratic test was used to determine the significance between treatment means ($P < 0.05$). Laying hens fed a graded level of MQ considerably enhanced ($p < 0.05$) egg production, feed intake, and egg weight at the end of week 12. In addition, layers diet supplemented with MQ showed increased ($p < 0.05$) haugh unit (HU), eggshell strength, and eggshell thickness at weeks 8 and 12. Moreover, HQ supplementation reduces the Aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), and cholesterol levels in layers. In conclusion, the current findings suggest that the addition of 0.06% of MQ supplementation in the laying hens' diet would be beneficial to enhance their egg production performance and certain aspects of blood profile parameters.

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Dietary incorporation of Zinc-Beta glucan improves immune status, gut microbial composition, egg quality, and mitigates gas emissions in laying hens' diets

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Beta-glucan has a variety of biological activities, including immune role enhancement, anti-infection, and glucose regulation. Previous studies reported that β -glucan supplementation has boosted immune cell activation in broilers' gut. Besides, some reports showed that supplementation of zinc could alleviate the negative effects of egg quality in laying hens. However, information regarding the positive effects of adding Zinc-Beta glucan additive on laying hen's performance is still unknown. Therefore, this study aims to explore the effects of dietary administration of zinc-beta glucan (ZB) on productivity, nutrient digestibility, gas emissions, blood profile, and microbiome analysis in laying hens' diets. A total of 192 Hy-line brown laying hens (1918.5 ± 0.52 g) were assigned to 4 distinct treatments for 12 weeks: TRT1, basal diet (control); TRT2, basal diet + 0.25% antibiotic; TRT3, basal diet + 0.05% ZB; TRT4, basal diet + 0.1% ZB. The addition of ZB (0.1%) to the hen diets resulted in increased ($P < 0.05$) egg weight, eggshell strength, eggshell thickness, and lymphocyte levels in the blood. The addition of ZB improved ($P < 0.05$) alpha and beta diversity in laying hens. Furthermore, taxonomic distribution analysis showed highly abundant phyla Firmicutes and Bacteroidetes and reduced ($P < 0.05$) pathogenic bacteria in ZB group layers. The findings suggest that ZB may enhance the immune status and egg quality while reducing gas emissions in laying hens by altering the gut microbial composition.

The supplementation of a mixed- phytase and xylanase product on older laying hens for improved phosphorus retention and ileal phosphorus digestibility

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Laying hens require phosphorus (P) for various biological functions, often supplied in excess as inorganic forms in diets. This practice, though inefficient and contributing to water pollution, can be addressed by incorporating enzymes in poultry diets. Enzymes enhance the availability of phytate-bound P in plant-based poultry diets, reducing the need for inorganic P. This study examined the effect of phytase and xylanase on P digestibility in laying hens. Novogen Brown Classic layers (n=314) were fed a control diet (T1) or a diet with an added with both phytase and xylanase (T2). Diets were provided for 12 days after which the excreta from each pen were collected for an additional three days to determine total retention and ileal digestibility of P and gross energy. Productivity parameters were also recorded over the course of the study. The amount of P detected in the excreta of the control group was higher ($P<0.05$) than the amount detected in the enzyme supplemented group. Both P retention and P ileal digestibility were higher ($P<0.001$) in the enzyme supplemented birds. Gross energy digestibility was higher ($P<0.001$) in the enzyme supplemented birds for both the total tract and ileal samples. There were only numerical improvements, not significant, in the layer productivity parameters. This study showed that the enzyme supplement can improve total retention and ileal digestibility of P and gross energy in older layers.

The effect of different calcium sources on eggshell mineral content and breaking strength in laying hens

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According to a 2014 study, 10-15% of eggs are lost due to eggshell quality before or during egg collection. An insufficient mineralisation (e.g. calcium) during the period of eggshell formation is speculated to be associated with poor eggshell quality. In this study, the effect of different blends of marine mineral complex (MMC) and limestone fed to laying hens were compared for mineral content and breaking strength of eggshell. A total of 448 hens (Species; Hy-line brown, 40 weeks old) were divided in a completely randomised design into 6 treatment groups with 74/76 hens per treatment. Six treatments were formulated with basic laying hens feed containing feed grade limestone (control; T1), or Coarse MMC (low dose; T2), Coarse MMC (high dose; T3), commercial MMC (T4), fine MMC (low dose; T5) and fine MMC (High dose; T6). The treatments had a significant effect ($p<0.05$) on breaking strength at week 4, 12, and 16. Breaking strength of eggshells from T4 group was 43N, 41N, 40N and 37N, while T1 was 41N, 37N, 37N and 36N at week 4, 12, 16 and 20 respectively. The treatments showed no significant effect on calcium and magnesium, but significantly ($p<0.05$) increased phosphorus (P) and Strontium (Sr) content of the eggshells. T2, T3, T4, T5 and T6 improved Sr content of the eggshells by 10%, 15%, 22%, 15%, and 20%, respectively and P content by 7%, 4%, 6%, 10% and 10%, respectively. Overall, the data demonstrates that the inclusion of MMC in laying hen diets improved breaking strength and mineralisation of eggshells.

Dietary Arginine Supplementation as a Mitigative Strategy Against Mycotoxin-Induced Adverse Effects in Laying Hens

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This study investigates the impact of dietary arginine (Arg) supplementation on counteracting the adverse effects induced by mycotoxins in laying hens. The research involved 240 laying hens, each with an initial average body weight of 1.66 ± 0.06 kg. These hens were divided into four groups, with six replicates per group and ten hens per replicate over a period of four weeks. The dietary treatments consisted of a non-toxin control diet containing 1.1% Arg (NT1.1) and mycotoxin-exposed diets supplemented with varying Arg levels: 1.1% (TX1.1), 1.2% (TX1.2), and 1.3% (TX1.3). The findings revealed no significant differences in egg production parameters such as egg weight, egg mass, and feed intake among the groups. However, a trend towards increased egg mass was observed in the NT1.1 group compared to the TX1.1 group. Similarly, there were no differences in shell color, thickness, hardness, albumin height, or Haugh unit, although yolk color tended to be higher in the NT1.1 group. Organ weight analyses showed no differences for the heart, spleen, and kidney, but a tendency for reduced liver weight was noted in the NT1.1 group. Additionally, no significant changes were observed in villus height or crypt depth in the jejunum and ileum, though the NT1.1 group showed a tendency for increased duodenal villus height. Toxin exposure led to up-regulation of interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF- α) in liver tissue. Similarly, interferon-gamma (INF- γ) and toll-like receptor-4 (TLR-4) gene expressions were elevated in the liver of the TX1.1 group. Muscle tissue analysis indicated increased IL-4 expression in toxin-exposed groups, with IL-6 and INF- γ also showing heightened levels in the TX1.1 and TX1.3 groups and the TX1.1 and TX1.2 groups, respectively. TLR-4 expression was specifically up-regulated in the TX1.2 group muscle tissue. The TNF- α expression significantly increased in the jejunum of the TX1.1 group compared to both the NT1.1 and TX1.2 groups. In conclusion, dietary supplementation with Arg appeared to mitigate liver damage and enhance the immune response in laying hens exposed to mycotoxins.

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Evaluation of the Efficacy of a Yeast Mannan Product on the Performance of Laying Hens

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Animal feed additives containing the mannan-rich fraction (MRF) extracted from the cell wall of *Saccharomyces cerevisiae* have been shown to improve productivity in various production animals. Productivity gains have been attributed to the ability of MRF to bind pathogenic microorganisms. As a result, including MRF in a layer diet can increase the microbiome diversity, helping maintain gastrointestinal integrity and improving nutrient utilisation. A feeding trial was designed to understand further the effects of MRF supplementation on layer hen productivity. A total of 256 28-week-old laying hens were allocated to one of 64 cages, and each treatment was replicated 32 times. Each cage contained four birds. The birds were fed either a diet with MRF (T1) or a negative control diet (T2). For birds fed the T1 diet, the MRF was fed at 800 g/tonne from 28 to 34 weeks of age and 400 g/tonne after 34 weeks of age. Observations included body weight, feed intake, egg production and quality and feed conversion efficiency (FCE). General health and mortality were also monitored daily. During weeks 0 to 4, 4 to 8 and 0 to 24, total egg weight (g/cage), egg numbers, egg mass, egg production and FCE were significantly greater in birds fed the T1 diet than in the T2 control diet. There were no significant differences between T1 and T2 diets in the bird live weight at the end of the experimental period. No changes in the bird's live weight between the start and end of the trial were observed. There were no significant differences between T1 and T2 diets in the percentage of cracked, soft-shell or dirty eggs. At day 168 of the study (end of the study), shell thickness was significantly greater in the T1 diets relative to the T2 control diet, and yolk colour was greater in the T2 control diet relative to the T1 diet. These results indicate that including MRF in standard commercial diets can improve a variety of layer hen productivity parameters.

Overview of heat stress management in fattening pigs: impact, influencing factors, and control strategies

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Heat stress in livestock production has gained significant importance in the last decade in Europe, mainly due to more frequent extreme weather conditions such as heatwaves resulting from climate change. These environmental conditions impact the welfare, health, and performance of livestock. Pigs are particularly sensitive to higher ambient temperatures due to their poor ability to sweat and pant. With only a few functional sweat glands and a subcutaneous adipose tissue layer hindering heat dissipation, pigs primarily rely on respiratory heat loss for thermoregulation. During the talk, an overview will be given of the various factors that influence the thermoregulation in pigs such as body weight, physiological status, genetics, climate, and environmental conditions. Next, we review the effects of heat stress on behavior, physiology, and performance of fattening pigs. Finally, we present the results of heat stress-reducing measures in pigs based on studies conducted during the last 3.5 years, including feeding strategies, management measures (e.g. stocking density and boar-line) and climate adaptations (e.g. increasing air movement and high-pressure fogging). These studies were conducted in the framework of the Vlaio LA project Coolpigs titled 'Heat plan for the Flemish pig industry' (project number HBC.2019.2877).

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Theatre 2

Antioxidant-enriched or energy-dense diet in heat stressed fattening pigs

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Antioxidants are considered potential mitigators of heat stress-induced damage. Additionally, modifying feed composition, specifically lowering crude protein (CP), aims to reduce internal metabolic heat production. This study investigated the impact of an antioxidant-enriched or energy-dense (E-dense) diet on heat-stressed fattening pigs. 192 mixed-sex fattening pigs were randomly assigned to a control, an enriched, or an E-dense diet. Dietary treatments were administered (ad libitum) when pigs reached ± 80 kg. The control diet comprised 15% CP, 3.6% crude fat, 0.4 mg/kg inorganic selenium (Se), and 100 ppm vit E; the enriched diet contained the same chemical composition but was supplemented with 0.2 mg/kg inorganic Se, 0.2 mg/kg selenomethionine, 200 ppm vit E, 200 ppm vit C and 0.1% betaine; the E-dense diet featured 13.6% CP, 6.6% crude fat, and increased energy and lys content. A one-week heat wave was induced when pigs were 20 and 22 weeks old. Antioxidant-enrichment of the diet resulted in a tendency to increased daily gain over the 6-week trial compared to the control group (891 vs. 925 g/day, $P = 0.090$). Over the entire trial, the E-dense group had a higher feed conversion ratio than the control group (2.95 vs. 2.67, $P = 0.006$). Carcass traits revealed increased fat thickness of 0.9 mm in the E-dense group ($P = 0.035$), along with lower lean meat content of 1.1% ($P = 0.002$). The meat of the enriched group displayed elevated vitamin E and Se levels ($P < 0.001$). Overall, the nutritional strategies did not significantly prevent physiological heat stress parameters, but supplementation of antioxidants could slightly ameliorate daily gain over the entire trial.

A dietary phytogetic solution mitigates heat stress indicators in pigs

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This study aims to corroborate if enhancements in performance and reductions in body temperature in heat-stressed (HS) pigs, supplemented with a 0.2% Capsicum spp.-based phytogetic solution (PHY) (Morales et al., 2023), are associated with changes in thermal tolerance markers. Forty-two pigs were assigned to three treatments: thermoneutral pigs on a control diet (TN-C), and pigs subjected to HS fed either PHY-supplemented (HS-PHY) or control diets (HS-C). After a 8-day experimental period, serum antioxidant enzyme activity, intestinal histomorphology, gene expression of HSP90 in the liver and muscle, and myosin in muscle were assessed. Superoxide dismutase activity was lower in HS-C pigs vs. TN-C and HS-PHY pigs ($P < 0.05$), being similar between TN-C and HS-PHY groups. Catalase activity was similar in TN-C and HS-C pigs, but higher in HS-PHY pigs compared to HS-C pigs ($P < 0.05$). HS-C pigs had shorter jejunum villi and deeper crypts than both HS-PHY and TN-C ($P < 0.01$). Expression of HSP90 in the liver was tentatively higher in both HS-C and HS-PHY pigs compared to TN-C pigs ($P > 0.10$). In muscle, HSP90 expression was significantly higher in HS-C pigs than in both TN-C and HS-PHY pigs ($P < 0.05$), but similar between TN-C and HS-PHY pigs ($P > 0.10$). Muscle myosin expression was lower in HS-C compared to TN-C ($P < 0.05$), with no difference between HS-PHY and TN-C pigs ($P > 0.10$). In conclusion, dietary PHY supplementation enhanced anti-oxidant enzyme activity, reduced HSP90 expression in Longissimus dorsi muscle, and elevated jejunal villus height compared to HS-C pigs, reaching levels akin to TN-C pigs, explaining the restored growth previously observed in HS-PHY pigs.

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Maternal supplementation with betaine or amino acids during lactation improves the viability of Iberian piglets under heat stress

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Lactating sows are highly vulnerable to heat stress due to the elevated metabolic heat generated during milk production. This can result in decreased feed intake, lower milk yield, impaired reproductive performance and a slowed piglet growth rate. The effects of supplementing lactating Iberian sows from day 70th of pregnancy onwards with betaine (BET; $n=16$) or amino acids (AA; $n=15$; arginine and valine) as nutritional strategies to mitigate heat stress was evaluated against a non-supplemented group (C; $n=16$). Average maximum and minimum temperatures were 32.3 and 23.4 °C, respectively. At birth and weaning, body weight (BBW and WBW, respectively), abdominal (AC) and thoracic circumference (TC) and total length (L) of each piglet were recorded. The BBW was higher in C piglets compared to AA and BET groups ($p < 0.001$). Moreover, piglets from the C group were longer and had larger AC than piglets from treated sows ($p < 0.05$). However, despite starting from litters with lower BBW than in the C group, no WBW differences were observed between groups at weaning, and AA piglets showed higher AC and TC than C piglets ($p < 0.05$). When using BBW as covariate, piglets body gain during lactation tended to be higher in AA and BET pigs compared to C pigs ($p = 0.070$). No significant differences on sow body weight changes during lactation were detected between groups. These results could indicate beneficial effects of supplementing lactating sows with amino acids or betaine. Further studies are required to confirm these results. Funded by Spanish Ministry of Science and Innovation (PID2021-125059OB-I00).

Heat stress effects on the gut microbiome profile of Iberian purebred and Duroc x Iberian crossbred weaned piglets

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Heat stress (HS) can adversely affect swine gut health by altering the microbiota profile, influencing the host's stress response, which can be different according to distinct genetic backgrounds. We investigated the impact of HS on the gut microbiome of weaned piglets from two distinct genotypes: Iberian purebred (IB) and Duroc x Iberian crossbred (DUIB). Forty pigs (20 per genotype) were individually housed in 20 pens and exposed to thermoneutral conditions (22°C) for one week, followed by a week under HS conditions (30°C), in two consecutive batches. Fecal samples were collected before HS (T0), two days after (T2), and seven days after HS (T7). Sequencing of the 16S rRNA gene and subsequent bioinformatic analyses showed significant differences in alpha and beta diversity across different time points and genotypes. Differential abundance analyses revealed one genus differentially abundant between T0 vs T2, 14 genera between T0 vs T7 and 17 genera between genetic backgrounds. Particularly, DUIB pigs exhibited a higher abundance of *Clostridium_sensu_stricto_1* compared to IB, only under HS, indicating a distinct HS response. The findings suggest a potential higher resilience to HS of the gut microbiome in IB compared to DUIB pigs. This work has received funding from the European Union's Horizon Europe research and innovation programme under the grant agreement No 01059609 (Re-Livestock project).

Maternal resveratrol improves sow's performance and the intestinal health and weight gain of suckling piglets under high summer temperature

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High summer temperatures affect the reproductive performance of sows. The present study aimed to investigate the effects of dietary resveratrol supplementation under high summer temperature on the reproductive performance of sows. Sows were fed either a control diet or a diet with 300mg/kg resveratrol during gestation and lactation. The results showed resveratrol supplementation increased litter weight. Resveratrol supplementation increased the abundance of IgA, IgG, and IgM in colostrum and intestinal sIgA levels of suckling piglets. Furthermore, we found that maternal resveratrol improved suckling piglets' intestinal morphology and affected the properties of exosomes-derived miRNAs in sow colostrum, as well as the genes targeted by differentially produced miRNAs. miRNAs are concentrated in the tight junction pathway, and the expression of intestinal tight junction proteins in suckling piglets increased. Our correlation analysis demonstrated the positive associations among significantly differential intestinal microbiota, intestinal sIgA production and SCFA concentrations, as well as the positive relation between the total daily weight gain and intestinal health of suckling piglets. Taken together, our findings indicated that maternal resveratrol supplementation could enhance the growth performance and intestinal health of piglets in a high temperature environment, which might be associated with the immunoglobulin and exosomes-derived miRNA in colostrum of sows.

Thermal adaptation in chicks: epigenetic regulation of resilience and vulnerability

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Stressful events during the embryonic or early life periods in chicks might lead to stress resilience or vulnerability throughout life and might be transmitted to the next generation. Here we demonstrate that heat stress during the critical developmental period of thermal-control establishment affects both body temperature and the expression of CRH in the hypothalamic paraventricular nucleus, later in life. CRH and body temperature increased during the heat challenge in chicks trained to be vulnerable to heat, whereas they decreased in chicks that were trained to be resilient. Accordingly, DNA CpG methylation (5mC) and hydroxymethylation (5hmC) at the CRH intron, which we found to serve as a repressor element, displayed low 5mC% alongside high 5hmC% in resilient chicks, and vice versa in vulnerable ones. In a second effort, we addressed the question of whether behavioral traits are heritable. A major obstacle in demonstrating transgenerational inheritance in mammals originates from the maternal environment's effect on offspring phenotype. To overcome this challenge, we used ovo embryonic heat conditioning of first-generation chicks and assessed the effect on their untreated offspring. We demonstrate heredity of both heat and immunological resilience, confirmed by a reduced fibril response in untreated offspring to either heat or LPS challenge. Furthermore, we demonstrate an association between epigenetic mechanisms and trait heritability, by genome-wide DNA-methylation analysis in the anterior preoptic hypothalamus of untreated offspring. Finally, we demonstrate a possible role for chromatin architecture in epigenetic heritability. Conclusively, we suggest a multilevel epigenetic regulation for stress responses and trait heredity.

Trend analyses revealed increasing heat stress situations in German regions with intensive turkey production

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Climate change can affect performance and health of livestock through the development of heat stress (HS). This study analysed trends of HS in German regions of dense turkey populations using enthalpy (h) values calculated from weather data of the past 50 years. Data from 15 weather stations provided by the German Weather Service were collected in the summer period and analysed retrospectively. Data contained the daily h maximum which was categorised to a HS class for poultry (HS0: <50 kJ/kg = no HS, HS1: 50-<58 kJ/kg = mild HS, HS2: 58-<67 kJ/kg = moderate HS, HS3: ≥67 kJ/kg = severe and extreme HS). For each region, the maximum daily h values achieved were assigned to the HS classes and their frequencies were determined. Trend lines for HS were created and their gradients specified. Finally, significant increases in the HS trend line gradients over 50 years were analysed (Mann-Kendall test). At all locations, h values between 11.8 and 83.6 kJ/kg were recorded. Classification into HS levels over all locations and years revealed the following frequencies: 78.7% of h values were assigned to HS0, 15.5% to HS1, 5.2% to HS2 and 0.6% to HS3. The gradient of all trend lines (over all regions and years) for all HS classes ≥ HS1 was positive and all gradients of the respective trend lines in the HS classes were significant. The presented study showed that the trend towards periods without HS (HS0) led to fewer days from year to year. However, in the HS classes ≥ HS1 with at least moderate HS, a significant increase of days with HS inducing h values was predicted. Thus, it can be expected that turkey production will face greater challenges in coping with HS in future.

The potential of in ovo-fed amino acids to alleviate the detrimental effects of heat stress on broiler chickens
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This study aimed to investigate the effect of in ovo-fed L-Leucine, L-Methionine, L-Cysteine, and their combinations on the performance, body temperature, and oxidative stress parameters of finishing broilers kept under heat stress (HS) or thermoneutral (TN) conditions. Treatments included a control (52 µL of sterile diluent/egg), T1 (1 mg of L-leucine/egg), T2 (3 mg of L-Methionine + 2 mg of L-Cysteine/egg), and T3 (0.4 mg of L-leucine + 1.6 mg of L-Methionine + 1.6 mg of L-Cysteine/egg). Treatments were in ovo fed through injection into the amniotic sac on embryonic day 18 (52 µL/egg). T3 increased ($P < 0.05$) body weight (BW) and decreased ($P < 0.05$) feed conversion ratio during the starter phase. Interestingly, males, but not females, from the in ovo-fed groups exhibited higher BW ($P < 0.05$) during the grower phase compared to the control group. During the HS phase, T2 and T3 decreased ($P < 0.05$) birds' facial temperature. Moreover, they were also the least affected by HS regarding feed intake and mortality. However, the BW of heat-stressed birds was not influenced by the treatments. In addition, in ovo-fed AA reduced ($P < 0.05$) lipid peroxidation products in both plasma and liver samples under both TN and HS conditions compared to the control group. In conclusion, in ovo-fed amino acids have the potential to improve thermotolerance, reduce oxidative damage, and alleviate the impact of heat stress on the performance of finishing broiler chickens.

Impacts of betaine and feed restriction on blood parameters in heat-stressed laying hens

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Poultry are highly sensitive to heat stress (HS) due to their insulating feathers and lack of sweat glands. HS could negatively affect the performance, egg quality, and health of laying hens. We studied betaine supplementation (BET) and/or applying feed restriction (FR) to mitigate the effects on blood parameters. We distributed 448 brown and 448 white hens over 4 treatments in 128 cages ($n=16$), during 6 weeks. All hens were reared under thermoneutral (TN) conditions for 21 days, followed by cyclic HS conditions ($32^{\circ}\text{C}\pm 2^{\circ}\text{C}$; RH 45–55%; 6h daily). The first week was considered acute heat stress (AHS) followed by chronic heat stress (CHS). Treatments (T1-4) were only provided during HS and were applied as a 2×2 factorial design with FR and BET. FR was applied for 4h in T1 and BET was supplemented in the drinking water in T2 (0.55 g/L). T3 received a combination of FR and BET and the control T4 was given normal drinking water and a standard diet. Blood was collected from 1 hen/replicate on days 11 (TN), 25 (AHS) and 39 (CHS). $p\text{CO}_2$ was significantly decreased during AHS and CHS in both laying hen breeds. Additionally, base excess and HCO_3^- were reduced and remained low until the end of HS. Interestingly, pH and $i\text{Ca}$ were not affected during AHS and CHS. Glucose initially increased during AHS but normalized during CHS. Na did not change and K only decreased in brown hens; hematocrit only decreased in white hens. Hyperventilation resulted in decreased $p\text{CO}_2$ with compensatory decreased HCO_3^- . pH and $i\text{Ca}$ were not affected and would emphasize the buffering mechanisms preventing alkalosis. Our results indicate that HS was experienced, however, metabolic responses were mild and not affected by BET or FR.

Impact of on-farm temperature on feed intake in Duroc pigs

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Although it is necessary to take heat stress caused by the feeding environment into account when genetic analyzing feed intake of pigs, there are few reports showing the relationship between feed intake and on-farm heat environment in field data. The aim of this study was to reveal the relationship between the daily maximum temperature in pig barn and daily feed intake in test period. The data was consisted of 17,757 records from 259 male Duroc pigs. The maximum daily temperature in the pig barns was calculated from the data installed in the pig barns. The daily maximum temperatures ranged from 21°C to 38°C. Using the daily maximum temperatures from the day of feed intake measurement up to 9 days before that, the plateau-linear regression model including first and second order age of pigs as covariate was used to estimate the fitness of the model by comparing the fit of the data for each day. In addition, the parameters of the best plateau-linear regression model were estimated. The results showed that the maximum daily temperature in the pig barn on the day of the feed intake measurement was better fitted by the plateau-linear model than the other days. The regression model indicated that daily feed intake decreased at temperatures above 24.9°C, decreasing by 49 g for every 1°C. These results indicate that the on-farm heat environment of pigs is an important factor when genetic analyzing feed intake, and that using the maximum daily temperature in the pig barn on the day of measurement in the plateau-linear model makes it possible to take these factors into account in the analysis.

Session 21

Poster 12

Effects of mineral interactions on oxidative stress in lactating sows subjected to heat stress

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This study investigated the effects of dietary minerals at different levels on reproductive performance, stress level, inflammatory response, antioxidant status, milk composition, and intestinal microbiota of lactating sows during high ambient temperature. The sows were divided into eight treatments according to a 2×4 factorial arrangement including two levels of Zn and Cu (ZC100, 100 mg/kg of Zn, and 20 mg/kg Cu; ZC150, 150 mg/kg of Zn and 25 mg/kg Cu) and four levels of Fe, Mn, and Se (FMS; NRC recommendation; FMS150, 150% of NRC recommendation; FMS200, 200% of NRC recommendation; and FMS300, 300% of NRC recommendation). A lower loss of both BW and backfat during lactation was observed in the FMS300 and FMS200 compared with NRC and FMS150. An increase in piglet survivability was observed in the FMS200 group compared to the FMS150 and NRC groups. Total superoxide dismutase (SOD) and Mn-SOD were increased in the FMS200 and FMS300 compared with the FMS. An increase in Se, Mn, and Fe levels in the milk was observed in the FMS200 and FMS300 compared with the FMS. The serum composition showed higher Se, Mn, and Fe levels in the FMS200 and FMS300 compared with the FMS. Conversely, a decrease in serum Zn levels was observed in the FMS200 and FMS300 compared with the FMS, while the Cu levels remained unaffected. The abundance of Proteobacteria phylum was significantly higher in the NRC group than in the FMS300 group. A higher abundance of Clostridium_sensu_stricto_1 genus was found in the NRC treatment compared to FMS300. In conclusion, increased Fe, Mn, and Se in sow diets improved weight loss and raised antioxidant levels. Yet, caution is warranted due to potential adverse effects on serum and milk Zn, which could impact litter growth.

Effect of heat stress on genetic evaluations for maternal traits in Iberian pigs

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The effect of heat stress on the reproductive performance of the Iberian sow can affect traits such as the farrowing rate or the weaning-mating interval. Since 2011, a selection program for Iberian pigs has been running, aiming to produce dry-cured products of supreme quality, which includes a maternal genetic index based on the number of piglets born alive (NBA) and litter weight at 21 days of age (LW21). The objective of this work has been to verify whether the high temperatures affect NBA and LW21. The Temperature-Humidity index (THI) has been calculated in 18,312 litters born from 2011 to 2024, at the mating day, days 23, 46, 69 and 92 after mating, farrowing day, and days 10 and 21 of lactation. THI was calculated using the average (THI_a) and the maximum (THI_m) daily temperature and humidity. Data were analyzed with models that included the type of the boar (Duroc or Iberian), the parity order (1 to 6) and the THI value, grouped into 18 (<9 to >24) or 25 (<11 to >33) levels for THI_a and THI_m, respectively. The results showed that for NBA there was no specific pattern for high THI values, in any of the days analyzed. However, for LW21 there was a decrease in performance at elevated temperatures, whether with THI_a or THI_m, with the best fitting model being THI_a at farrowing day. The grouping of the data into a fixed effect with two levels (THI_a<21 and THI_a>20), and its inclusion in the BLUP-animal genetic evaluation model, resulted in an estimate of -0.986 kg for THI_a>20. In conclusion, for future genetic evaluation a climate adjustment is not necessary for NBA, but it is for LW21, applying specific models for each trait.

Heat stress effects on the growth and metabolism profile of Iberian purebred and Duroc x Iberian crossbred weaned piglets

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Heat stress (HS) is a major concern in pig industry. Rustic breeds like the Iberian pig are more resilient to HS than commercial breeds. We investigated the impact of HS on the metabolic profile and enzyme concentration in Iberian purebred (IB) and Duroc x Iberian crossbred (DUIB). Forty pigs (20 per each genotype) were individually housed in 20 pens and exposed to thermoneutral conditions (22°C; TN) for one week, followed by a week under HS (30°C), in two consecutive batches. Weight was measured at the beginning and ending of the TN period and at the end of HS period to calculate average daily weight gain (ADWG), fractional growth rate (FGR) and feed conversion ratio (FCR). Blood samples were collected before HS (T0) and two days (T2), and seven days (T7) after the start of HS in the morning and the afternoon. FGR was higher during HS than TN, but increased more in IB than DUIB pigs. Glucose metabolism decreased in T2, while lipidic metabolism in IB pigs and protein metabolism in DUIB pigs were increased in T7. Enzymes related to tissue damaging increased more with HS in IB than DUIB pigs possibly related to the higher oxidative stress status previously studied in the IB pig. Thus, HS stress affected both IB and DUIB animals' metabolism and stress but differently between breeds. This work has received funding from the European Union's Horizon Europe research and innovation program under the grant agreement N°01059609 (Re-Livestock project).

Heat tolerance in Polish local pigs: preliminary results of metabolomics analysis of blood

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The heat tolerance becomes an urgent issue in livestock farming. It is assumed that the local breeds have a higher resilience to the rapid changes in the temperature, especially the heat waves. Here we evaluate the blood parameters (standard and metabolomic) in Polish local pig breed. In total 23 sows and boars were used: 11 females from Złotnicka White breed with body weight ~140kg, 6 females from Złotnicka White x Złotnicka Spotted cross (body weight ~150kg), and 6 males came from Złotnicka White x Złotnicka Spotted cross (body weight ~150kg). The pigs were kept on a small farm in Poland (52°36'50"N 17°20'30"E) and fed standard commercial diet for fatteners with unlimited access to water. The barn did not have controlled environment. The blood was collected by the veterinarian on the evening of 19th June and morning of 20th June 2023. Frozen plasma was sent to Metabolom USA for metabolomic analysis (data to be obtained). The statistical analysis of the blood parameters was done in R statistical package. The significant differences between morning and evening samples were observed only for free fatty acids and eosinophils levels. Between breeds the differences were significant for alanine aminotransferase, aspartate aminotransferase, magnesium, percentage of lymphocytes, erythrocytes, hematocrit, hemoglobin, hemoglobin concentration. In case of sexes the differences were between aspartate aminotransferase, alkaline phosphatase erythrocytes, hematocrit, hemoglobin, hemoglobin concentration. Further investigation is needed to evaluate those results and correlate them with metabolomics. This project was financed by the European Union grant no. 101059609.

Session 22

Theatre 1

Breed Differences in Semen Quality Characteristics in Commercial Boars

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This study investigated differences in semen quality parameters in 6 commercial pig breeds. Boar age, rest days between collection, progressive motility (PM), motility (MO), concentration (CO), and volume (VOL) from CASA data was recorded on 3908 boars from 6 different breeds (Duroc=1731, Hampshire=158, Landrace=693, Large White=269, Pietrain=212 and Synthetic Line =845) between January and June 2023. Boars were housed in 30 boar studs from 9 countries. 20 semen collections per boar were needed for inclusion in the study yielding a total of 94372 records. Data was analysed using mixed model equations including semen quality characteristics as predicted variables, breed, collection weekday, and collection month as fixed effects, boar age and rest days as covariates, linear and quadratic covariates and boar nested within stud as a random effect. Weighted least square means were calculated for unbalanced groups. Breed differences were observed for all semen parameters ($P < 0.05$). Lower PM ($77.4 \pm 2.07\%$), MO ($88.5 \pm 0.84\%$), CO (0.27 ± 0.025) and higher VOL (364.2 ± 26.33 ml) were observed for Hampshire pigs and they differed from the rest of the breeds ($P < 0.05$). PM differ between Landrace and Synthetic Line pigs while MO was higher for the Synthetic Line than for Landrace and Duroc pigs ($P < 0.05$). CO was higher for Duroc and Landrace pigs than for Large White and Synthetic Line pigs. VOL differed between all breeds except for Pietrain pigs. Results demonstrate differences in major parameters impacting semen production and quality which ultimately impacts the number of offspring an individual boar can influence. Our results highlight the need to continue to understand within and between breed differences as new breeding technologies are developed.

On the genetics of farrowing duration in pigs

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In the last decades, genetic progress for litter size in pigs has substantially evolved, and consequently, farrowing duration (FRD) has increased. Prolonged FRD is associated with a higher number of stillbirths and impacts the required labour per sow during farrowing and colostrum intake of the newborn piglets. FRD is a difficult-to-measure trait due to the high labour demand and, therefore, it is not commonly available. To our knowledge, most of the previous studies on FRD evaluated data on a few hundred sows and the genetic parameters for this trait have not yet been reported. The goal of this study was to investigate the genetic background of FRD using a large commercial dataset. The genetic parameters for FRD are reported here and a genome-wide association study is currently being carried out. The evaluated data consisted of 14902 litters of 4581 purebred Large White sows from one farm located in Brazil. The data was collected between 2020 and 2023 as part of the routine data collection of the farm that counts with employees in the farrowing room at all times. The time of birth of each piglet was individually recorded and the FRD was defined as the time between the birth of the first and the last piglet. The genetic parameters were estimated with an additive linear model using ASReml v3. The fixed effects were population mean, type of litter (purebred/crossbred), farrowing room, year-week, parity number of the sow, average birth weight of the piglets and litter size. The random permanent, additive and residual effects were also fitted in the model. The average FRD and litter size in this dataset were 216±110 minutes and 15.0±2.7 piglets, respectively. Population mean, year-week, parity number of the sow and litter size were the only significant fixed effects ($P < 0.001$). A heritability of 0.07±0.01 was estimated and the permanent effect accounted for 0.03±0.01 of the total phenotypic variance. A clear linear relationship between litter size and FRD was also observed. Although the heritability of FRD was low, this study brings new insights that may contribute to the genetic improvement of this trait.

Session 22

Theatre 3

Dam parity is associated with reproductive performance in primiparous sows

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This study aimed to investigate associations between early life reproductive performance indicators and the dam parity the female was born into. Data was collected between 2019 and 2023 in 11 European L03 PIC commercial multipliers. Lifetime performance records were available on 44909 females born from sows parity 1 (n=30144), 2 (n=10227), 3 (n=3320), and 4+(n=1218). Data included lifetime daily gain at off-test (c. 165.5 ± 20.53 days of age; LDG), age at first mating, age at successful mating, age at first farrowing, P1 total born (TB) and born alive (BA) piglets, P1 wean-to-service (WSI) interval and P1 non-productive days (NPD). Data was analysed using univariable generalised linear mixed models with dam parity included as fixed effect and contemporary group (i.e. farm, year and month when entering the herd) as random effect. A Gamma distribution was fitted to WSI and NPD. Females born from P1 sows had lower LDG, and higher age at first and successful mating, age at first farrowing and WSI ($P < 0.001$) and lower TB (-0.3 piglets) and BA piglets (-0.2 piglets; $P < 0.001$) when compared with other parity groups. NPD was similar between females born from P1 and P2 sows and higher to those born from later parity sows ($P < 0.001$). There were no observed differences ($P > 0.05$) between sows P2-P4+ in any of the other studied traits. Results suggest that selecting replacement gilts born from gilt litters could have a significant impact on reproductive performance variation and overall farm efficiency. As with gilts making up 25% of the breeding herd, selection of replacements from gilt litters is unavoidable., our results emphasise the need to implement targeted gilt development programs to maximise their productive life

Validation of a Piétrain sire breeding program on crossbred piglets' vitality and congenital defects

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Decreasing mortality and the prevalence of congenital defects in the farrowing house is important in pigs. Tackling these issues will lead to both economic gains and an improved animal welfare. We validated a Piétrain sire breeding program for piglet vitality and congenital defects using two datasets. First, the breeding organization collected data on 14186 crossbred litters (Piétrain sire, hybrid dam) on piglets' vitality (V), number of piglets born death (NBD), number of porcine congenital splay leg syndrome (PCS) and scrotal hernia (SH) piglets, and preweaning mortality (PWM). Second, a similar, independent test (4561 litters) was performed by ILVO (Flanders research institute for agriculture). Validations were done by correlating mean phenotypic values and breeding values for the 384 common boars between both datasets. Breeding values were estimated by a genetic sire-dam model with ssGBLUP and the blupf90 software. Heritabilities were low for V (9.5%), SH (12.2%), PCS (14.1%), PWM (13.8%) and NBD (5.5%). The phenotypical correlations were significantly >0 for V (rp=0.25) and PCS (rp=0.33), but not for SH (rp=0.06), PWM (rp=0.03) NBD (rp=-0.08). Correlations of breeding values were positive for V (rg=0.22), SH (rg=0.05), PCS (rg=0.47) and PWM (rg=0.40), but not for NBD (rg=-0.10). Our findings show that breeding Piétrain sires for vitality and congenital defects can improve vitality and decrease the prevalence of congenital defects in crossbred piglets. Further research is needed to optimize the program for survival traits.

Session 22

Theatre 5

Genetic trends in the lysine requirements of growing-finishing pigs and reproductive sows, due to genetic trends in production traits

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The requirements for SID lysine due to lean tissue growth rate (LTGR) in growing-finishing pigs were quantified based on data from the literature. The same was done for those requirements due to LTGR, due to protein deposition in mammary tissue, uterine tissue, fetal tissue and placental tissue, and due to colostrum and milk protein synthesis in gestating and lactating sows. These processes were expressed in terms of LTGR, litter size, piglet birth weight, preweaning survival rate, litter weaning weight and teat number. The 2013-2023 genetic trends of those traits in the PIC-L02, PIC-L03, PIC-L65 and PIC-L800 populations were estimated, and converted to genetic trends in lysine requirement. Based on this, the daily SID lysine requirement of Camborough (i.e. PIC-L02 × PIC-L03) sows increases by 0.47 g/d (i.e. by 2 % of the current mean level) per year: 0.10 g/d per year and 0.37 g/d per year for gestational and lactational processes, respectively. The daily SID lysine requirement of growing-finishing progeny of Camborough sows and either PIC-L65 or PIC-L800 boars increases by 0.09 g/d (i.e. by 0.6 % of the current mean level) per year.

Genetic Effects on Weight in Growing Piglets (prae iPig project)

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The prae iPig project aims to understand various aspects influencing growth, resilience, and health in sows and piglets. We looked at the genetic effects on weight in growing piglets. The weight of suckling piglets of 21 primiparous sows was recorded. Piglets had access to the sows feed in addition to creep feed. Piglets were weighed at birth and on days 3, 7, 14, 21, and 28. All sows and 94 piglets were genotyped with the PorcineSNP80v1_HTS_20033000_A1 SNP chip. Unmapped SNPs or SNPs on the X or Y chromosome were removed to reduce a loss of power due to multiple testing; SNPs with a GC score >0.7 and a SNP call-rate >0.9 were retained, leaving 47,745 SNPs. During quality control, 5 piglets were removed for various reasons. A mixed linear model was fitted to the data including the Weibull function to model piglet growth, and the sows used as a random effect. Variables with a significant effect on the weight data included: experiment (2 separate trials), piglet sex, litter size, and weekly sow weight. No significant SNP effects were found across test-days; but several regions of interest were identified when analyzing test-day data individually. Three regions reached Bonferroni corrected significance: on chromosome 2, 2 markers at 40,681,608 and 83,569,250 bp; on chromosome 6, two markers at 111,277,681 and 112,819,536 bp; and on chromosome 12, 5 markers between 13,818,106 – 32,527,645 bp. A larger region on chromosome 6 includes multiple consecutive markers with p-values <0.0001 for several test-days (bp-positions are based on 10.2 assembly).

Session 22

Theatre 7

Differentially expressed genes in Piétrain sired pigs when feeding a high-fat high-fibre diet

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The Piétrain is a Belgian pig sire breed that is primarily selected for superior carcass quality and efficient feed conversion, hence commonly fed nutrient-dense high-grain diets. However, at present, it is unclear how Piétrain-sired progeny might cope with byproduct-based feeds that are higher in fibre and fat compared to conventional feeds that are nutrient-dense high-starch. Therefore, we investigated the effect of a dietary shift on gene expression in hypothalamus, colon and ileum tissue of 15 week-old pigs. Thirty-two pigs were included in a 2x2x2 experiment with diet (control, CON, vs. high-fibre high-fat, HFF), sex (castrated male vs. female), and genetic background (high vs. low estimated breeding value (EBV) for feed intake) as factors. Pigs were fed with CON feed until 14 weeks, thereafter separated into two groups, and fed for one week until euthanasia on the CON feed (n=16) or the HFF feed (n=16). Quantseq was performed on total RNA samples followed by differentially expressed gene (DEG) analysis using DESeq2 in R. Comparing CON with HFF feed, 20 DEGs were identified in the hypothalamus, whereas for sex, 18 DEGs were detected in colon tissue and 19 in ileum tissue. When comparing low with high EBV for feed intake, 13 DEGs were found in colon tissue and 9 in ileum tissue. These results will provide insight into the genes that are involved in the regulation of the metabolism of byproduct-based feeds.

Sires of low ad libitum-residual feed intake progeny have no undesirable effects on growth performance, gain composition and carcass traits of restricted-fed heavy pigs

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We assessed the impact of sire classification for ad libitum residual feed intake (RFI) on feed efficiency, growth performance, tissue accretion, gain composition, carcass, and fat quality traits in restricted-fed heavy pigs. The experiment involved 420 purebred pigs from the Goland C21 sire line, offspring of 23 sires, which were fed ad libitum or restricted, and tested between 96 and 168 kg. RFI was estimated as the residual of a model accounting for empty body weight, body lipid and body protein mass, which were estimated from measures of body weight and backfat depth. The 23 sires were classified into low-RFI (LRFI), high-RFI (HRFI) or average-RFI (MRFI) based on their effects on RFI of the ad libitum-fed progeny. Ad libitum-fed offspring of LRFI sires showed increased feed efficiency and reduced feed intake, with no differences in gain composition or carcass traits. Restricted-fed progeny of LRFI sires exhibited reduced energy and feed intake while maintaining similar growth rates compared to HRFI group. Moreover, LRFI group showed slightly higher fat cuts % ($P < 0.1$), ham weight % ($P < 0.05$), and significantly higher ($P < 0.01$) ham subcutaneous fat quality (-4% iodine number, -8% linoleic acid, +5% stearic acid content) than HRFI group. Hence, a reduction in RFI is not expected to adversely affect growth performance, gain composition, carcass, and ham quality traits in restricted-fed heavy pigs.

Session 22

Theatre 9

Impact of selection for birth weight uniformity in meat quality traits in SRC Iberian pigs

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Since 2022 an experimental design with discrete generations and the first parturition of the sows is being performed in a Sánchez Romero Carvajal Iberian pig population. Generation 0 and 1 has been elapsed since then. The objective of this study was to analyse the data of this selection experiment focused on increase in the uniformity of piglet's birth weight (BW) and evaluate the impact in meat quality traits. Two replicates per generation have been performed. A total of 502 first litters belonging to nulliparous sows were used with a total of 3224 piglets BW. A heteroscedastic model was used to select animals, including mother age, piglet sex, litter size at birth and replicate as systematic effects. Genetic effects (mean and variability) were assigned to the mother. The litter effect was included as random effect. The pedigree contained 2699 animals. The following meat quality traits were studied in 187 sows: carcass, ham, weight, loin weights, loin intramuscular fat content and backfat composition of 12 fatty acids. The genetic coefficient of variation on BW was 0.56, with a genetic correlation between birth weight and its residual variance of -0.27, but not significantly different from 0. Regarding quality traits, the correlations with predicted breeding values for birth weight environmental variance ranged between 0.21 (daily gain in Montanera and loin weight) to -0.13 (Oleic + Linoleic, %). First results on generation 0 indicates that none of the quality traits routinely measured will be negatively affected by the selection towards BW uniformity

Differential Cold Adaptation Mechanisms: Adipose Tissue Subpopulation Variances in Tibetan and Bama Pigs
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Thermogenic beige adipocyte displays a remarkable ability in mammals to adapt to cold environments, but the underlying cellular mechanisms remain unclear, especially in pigs that lack functional UCP1. Here, we observed a high number of beige adipocytes in adipose tissues from Tibetan pigs (cold-tolerant) compared to Bama pigs (cold-sensitive) after short-term cold exposure (4 °C for 3 days). Through single-nucleus RNA sequencing of adipose tissues from both pig breeds at room temperature, we discovered two cell subpopulations that were specific to Tibetan pigs, PDGFR α +EBF2^{High} and ADIPOQ+HIF1A^{High}. PDGFR α +EBF2^{High} cells were characterized as potential beige precursors, while ADIPOQ+HIF1A^{High} cells were found to highly express thermogenic-related genes. Despite the disappearance of the lipogenic subpopulation and the appearance of the lipolytic and the thermogenic subpopulations observed in both pig breeds upon cold treatment, Tibetan pigs exhibited stronger cellular and molecular responses compared to Bama pigs. Remarkably, cold-induced de novo beige adipogenesis and white adipocyte browning were observed in Tibetan pigs, while Bama pigs relied more heavily on white browning. Moreover, BMP7, which was highly expressed in PDGFR α +EBF2^{High} subpopulation, positively regulates porcine beige thermogenic capacity. Collectively, our data offers a comprehensive and unprecedented perspective on the heterogeneity and plasticity of adipose tissues of pigs and broadens the understanding of beige biology in mammals.

Exploring the potential of genetic selection for enhanced pork belly quality

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Pork belly accounts for ~15% of chilled carcass weight, nowadays often representing the most valuable primal cut in North America (\$/cwt). Packers and international buyers prefer fairly thick and firm bellies, as these have a higher processing yield and profitability potential. However, historic genetic selection towards lean carcasses has led to thinner pork bellies and firmness issues. While the potential to enhance certain pork belly dimensional traits has been reported, limited research has focused on quality traits, such as firmness. The multifactorial nature of belly firmness complicates selection for belly quality. However, genomic evaluation and selection could provide further opportunities. In this study, traditional and novel phenotypic traits were evaluated in carcasses from 1,500 commercially produced and genotyped crossbred pigs. Heritabilities for most belly specific traits were moderate to high, showing potential for genetic selection. Positive phenotypic and genetic correlations were observed between objective belly firmness measurements and fat content/thicknesses in both the loin and the belly primals, total belly thickness, subjective firmness scores, as well as backfat saturated fatty acid content. On the other hand, belly firmness was negatively correlated to carcass and individual primal lean meat yield, polyunsaturated fatty acid content, and backfat iodine value. Genome-wide association analysis revealed specific genomic regions with potential to enhance selection for belly quality traits. These results indicate that pork belly quality can be improved by including direct and correlated traits in selection objectives in pig breeding programs.

Sire selection for infrared prediction of ham weight-loss during dry-curing affects green ham traits, and measured ham weight-loss in heavy pigs

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Excess ham weight loss (WL) during dry-curing results in a reduction of sellable product and compromises the quality of dry-cured hams. Early predictions of 12-month WL (pWL), based on infrared spectroscopy, exist and are highly genetically correlated with measured WL. However, effects of selective breeding for pWL on measured WL and other quality traits have never been validated. This study aimed to assess the effects of selective breeding for pWL on carcass, green ham quality traits, and WL measured at 18 months of dry-curing in a selection experiment. Estimated breeding values (EBV) for pWL were obtained for 70 sires of the Goland C21 line using an animal model and BLUPF90. The 6 top (low pWL EBV) and 6 bottom (high pWL EBV) sires were selected for mating with PIC Camborough® sows and produced 349 pigs in total. Pigs were raised according to Protected Designation of Origin dry-cured ham specifications, and slaughtered at 9 months of age and 166.1 ± 15.9 kg body weight. Thighs were further processed for dry-curing. Compared to the progeny of the bottom sires, offspring of the top sires exhibited significantly lower WL after 12 and 18 months of dry-curing ($P < 0.05$), and increased ham subcutaneous fat depth ($P < 0.01$). In conclusion, selective breeding for pWL proved to be effective in reducing WL and increased ham quality in the subsequent generation.

Session 22

Poster 13

Association between feed efficiency traits and disease resistance genes in pigs

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It is possible to genetically enhance resistance to disease by selecting for genotypes of genes that are effective in suppressing disease. However, there are few reports on the association between disease resistance gene genotypes and feed efficiency in pig, and the impact of selection by these genes on feed efficiency has not been revealed. In this study, we examined the association between SNP polymorphisms of disease resistance genes and feed efficiency traits. Phenotypes were feed conversion ratio (FCR), average daily feed intake (ADFI), and average daily gain in test period (ADG) for 118 male Duroc pigs. Two SNPs in NOD1, one SNP in NOD2, and one SNP in NLRP3 were used as SNPs for disease-resistance genes. The effects of SNPs were tested using a linear model including the season at the end of the test (4 levels) and the genotype or additive and dominant effects of each SNP. Results showed that no SNPs had a significant effect on FCR and ADG. For ADFI, the genotypes and additive genetic effect of the SNP (G1992A) in NOD1 were significant ($P < 0.05$). This gene is involved in the recognition of peptidoglycan, a bacterial component, and individuals homozygous for the gain-of-function allele had significantly increased ADFI compared to heterozygous individuals. The results suggest that selection for disease-resistance genes has little negative effect on feed efficiency in pigs.

A study on the effect of not approved semen doses on fertility traits

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Large sets of data derived from thousands of individual sperm measurements are routinely provided by CASA (Computer Assisted Sperm Analysis) System in the Norwegian Artificial Insemination (AI) center to produce Duroc and Landrace semen doses. The assessment of the quality of an ejaculate is the first step in semen production to ensure high quality of the AI doses used in the customers' herds. It is of high importance to ensure that quality of ejaculates, since increased number of rejected ejaculates would lead to decreased efficiency at the AI station and loss of genetic progress. In this experiment, inseminations were conducted with 4092 approved and 448 not approved semen doses between April 2021 and December 2022 on purebred sows in ten different nucleus herds. Not approved semen presented somewhat lower thresholds for tail defects, proximal and distal droplets than what is usual in the production of AI doses. Motility was within normal range. The effect of approved or not approved semen (1/0) and three different morphology traits on field fertility traits (farrowing rate, total number born and stillbirth) were analyzed by using ASReml. Results shows no effect of approved or not approved semen and morphology traits on the fertility traits in field, except a small effect of distal droplets on farrowing rate for the Duroc breed. This circumstance allows considering utilizing semen doses of somewhat inferior quality for some of the morphology traits than what is conventionally deemed as standard.

Session 23

Theatre 1

Keel bone lesions in laying hens: occurrence and risk factors

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In Europe, farms for laying hens are housing the highest number of caged animals, whereas a transition towards cage-free systems is expected based on the Initiative of the European Citizens "End the Cage Age" and the corresponding Resolution of the European Parliament to provide hens with more possibility of movement, besides other improvements with respect to animal welfare. On the other side, navigation ability of laying hens in alternative systems, with special reference to multi-level aviary systems, may be associated to collisions with equipment and other animals which can produce injuries to animals where and if other risk factors are present. In fact, the latest Scientific Opinion of EFSA about the welfare of laying hens on farm has identified several highly-relevant welfare consequences for these animals based on severity, duration and occurrence, with bone lesions at the top of the list. Among these latter, keel bone fractures have been proved to produce negative states such as pain, distress, and/or discomfort, whereas less data are available on the occurrence of keel bone deviations and their impact on animal welfare and health. In this context, the present contribution aims to provide an overview of the available data about the occurrence of keel bone fractures and deviations in laying hens kept in different rearing systems, discussing the ontogenetic, management and housing factors that have been identified as risk factors, and, finally, to provide information about the animal-based indicators both on farm and slaughterhouse.

Effects of a Bacterial Compound on Litter to reduce Footpad Lesions in Broilers

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A trial was performed in broilers to reduce Footpad lesions (FPL) that often affects flocks' welfare. A total of 89,200 Ross308 chickens (39±3g) were housed in 2 sheds (C, control; T, treated) at the same environmental conditions. Females were housed in the first part of each shed, and males in the second and third part. A bacterial bedding conditioner (EazyBed Pro, Lallemand, France) was applied on litter in T. In C no treatments were carried out. Females were slaughtered at 36 days (T: 1528±195g; C: 1562±188g) and males utilized the whole space until 43 days (T: 2696±296g; C: 2737±364g). At 20 days and before capture, 30 birds living in each part of the shed were randomly selected and, from both pads, the perimeter shape of each lesion was recorded. Shape measurements were obtained in cm², using the imitoMeasure application (Imito AG, Swiss). For bacteriological and mycological culture, litter was sampled at different depths, pooling 9 samples/part, at the same timepoints. At slaughtering, 12 legs for each group were randomly selected for histological examination, characterizing the severity of lesions. At 20 days, FPL were not observed in birds. At 35 days, lesions measured in T (0.57 ± 0.08cm²) were significantly lower than in C (1.47±0.14cm²; p<0.0001). At 42 days, lesions measured in T (0.65±0.18cm²) were significantly lower than in C (1.76±0.34 cm²; p=0.049). In litter collected in T, Gram negative bacteria (p=0.0015) and Staphylococcus spp. (p=0.0386) significantly decreased (CFU/g), in particular in the second part of the shed (p=0.0098, p=0.0131 respectively). No significant differences were recorded for fungal flora. In T, broilers showed a total histological score significantly lower than in C (p=0.0002), in particular in males (p<0.0001). The use of the compound positively affects width and severity of FPL, supported by bacteriological analysis that showed a reduction of the total charge of bacteria commonly involved in the condition.

Session 23

Theatre 3

Effects of genotype and husbandry system on pododermatitis and plumage soiling in organic turkey production

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Effects of different genotypes and husbandry systems with 100% organic feeding on foot pad dermatitis (FPD) and soiling in turkeys were studied. Non-beaktrimmed male turkeys of a slow-growing genotype (Auburn (A), n=256) and a fast-growing genotype (B.U.T. 6 (B), n=128) were raised at two research stations indoors during rearing (8 weeks). Twelve pens were equipped either without environmental enrichment (EE) at station 1 (H1-, n=144) and with EE (pecking stones, elevated seating platforms, silage feeding) at station 2 (H2+, n=240). During the fattening period (9 to 20 weeks) half of the animals per pen of H2+ were relocated to a free-range system (H3-MS, n=104), while animals of H1- remained in the indoor system. Four-weekly, FPD was scored from 0 (=no damage) to 4 (=severe damage) and soiling at back, breast and butt from 0 (none) to 3 (severe). The high prevalence of FPD (51%) and soiling (breast area: 68.8%) was affected by genotype, husbandry and age (p<0.001). Genotype A, which was scored worse than B (p<0.001), showed the worst FPD in H3-MS (p<0.001). For soiling at breast, genotype B was scored worse than A (p<0.001). Investigated traits did not differ between free-range access and indoor systems. In conclusion, strategies to optimize free-range systems and selection for better footpad health in organic turkeys are needed.

Incomplete Milking for metabolic relief of dairy cows

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The metabolic challenge related to high milk production may be augmented by various stressors. We herein aimed to test whether temporally lowering milk yield (MY) by incomplete milking (IM) may reduce metabolic stress in multiparous Holstein cows during peak to mid lactation. The IM was implemented using a software-module which enabled automated early cluster removal according to an individual pre-defined amount of milk. In the 1st IM-phase, MY was gradually decreased for 5 d by 9%/d. In the 2nd IM-phase, the reduced level (~40%) was held constant for further 5 d. Before and after IM, IM cows (n=17) were milked equally as the CON cows (CON; n=18; milk flow-based cluster removal at <0.3 kg/min). Two wk before, during and 2 wk after IM, daily MY, fatty acid (FA) concentration in serum, cytobacteriological and other analyses in milk were assessed weekly. Linear mixed-models and t-tests were used for data analysis. The IM reduced daily milk withdrawal from 39.8±7.2 kg/d to 24.9±4.4 kg/d (~40% reduction) in the IM group, whereas, during the same period, MY in the CON group did not change (~38.0 kg/d). Due to lesser milk withdrawal, the low FA concentration in the IM group (0.17±0.14 mM) was further reduced to 0.07±0.04 mM (P<0.01) during IM, but FA concentrations in the CON group did not change (0.15±0.15 mM). However, somatic cell count remained on a low level in both groups (IM: ~26.3×10³ cells/mL, CON: ~33.3×10³ cells/mL). In the wk after IM, MY in the IM group (34.6±5.8 kg/d) tended to be less than in the CON group (38.3±6.9 kg/d; P=0.09). Thereafter, no difference in MY was observed. The IM applied lead to metabolic relief and may be considered for maintaining and improving health in dairy cows during stressful conditions such as ketosis or heat stress.

Session 23

Theatre 5

Influence of incomplete milking on the metabolic and oxidative stress status in early-lactating cows with high lipolysis

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To meet the high energy demands around calving, high-yielding cows mobilize body fat and can experience metabolic and oxidative stress. We hypothesized that temporary incomplete milking (IM) can positively affect the metabolism in early-lactating cows with high lipolysis. Multiparous Holstein cows (n=46) were allocated to a control (CON) or an IM group. Before and after the IM phase (week (wk) 2 and 3 postpartum (pp)), both groups were milked conventionally (cluster removal at <0.3 kg milk/min). During IM, the IM cows were milked up to a defined milk quantity (IM:~32 kg/d, CON:~11% more). Fatty Acids (FA), β-hydroxybutyrate (BHB), and Reactive Oxygen Metabolites (dROM) measured in serum were obtained weekly in the first 7 wk pp. However, the CON and IM group did not differ in their metabolic status. Based on their FA concentrations, we further classified cows within the first 2 wk pp: FA >800 μM (CON-high: n=5, IM-high: n=5) or FA <800 μM (CON-low: n=18, IM-low: n=18). Data (presented as mean±SE) were analyzed using mixed models and post hoc tests. Lesser FA values even at wk 4 pp in IM-high cows (374±88 μM) compared to CON-high cows (888±352 μM; P=0.02), indicate decreased lipolysis due to IM. The subsequent 1.7-fold increase of FA values at wk 5 pp in IM-high cows (P=0.03) may reflect the rising energy demand after the IM period. Compared to CON-high cows, IM-high cows had lower BHB values at wk 4 pp (IM-high: 632±76 μM; CON-high: 2,059±948 μM; P<0.01) and also at wk 5 and 6 pp. The dROM values in IM cows remained constant. In conclusion, the metabolic response of early-lactating cows to IM varies. Cows with elevated FA values seem to profit from the treatment.

Insights into Mastitis Management: Unveiling the Drivers and Goals Behind Swedish Dairy Farmers and Veterinarians Decision-Making

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Mastitis, a prevalent health issue in dairy farming, poses significant economic challenges for farmers and raises public health concerns related to antibiotic use for treatment. Previous studies have highlighted low adherence to veterinarian advice as a common issue. To address and control the situation farmers can adopt preventive animal health management measures to control disease incidence and to prevent further spread on the farm once identified. However, there is limited understanding of the reasoning influencing farmers' adoption of specific control measures and whether their choices differ from the advice and reasoning provided by veterinarians facing similar situations. In this study, we explore the drivers and motivations of both farmers and veterinarians, shedding light on the rationale behind their management behaviours aimed at reducing and controlling mastitis incidence on the farm and the goals driving these behaviours. Based on goal-system theory, our study design explores the motivational basis for the adoption of control measures when the somatic cell counts (SCC) exceed a certain endogenous reference point decided by the farmer. We further explore the advice veterinarians would provide in a comparable situation. For this, 30 randomly selected Swedish dairy farmers and 27 veterinarians were interviewed using the Laddering technique to unfold the stage-dependent goal-means chains when handling mastitis outbreaks. This study provides insights into the complexities faced by both farmers and veterinarians offering a deeper understanding of the motivations possibly influencing adherence to veterinarians' advice. It highlights the necessity of tailoring animal health recommendations to align with farmers' motivations, emphasizing the need for individualized approaches to handle mastitis on each farm.

Session 23

Theatre 7

Impact of early life experiences on the development of ear necrosis in weaned pigs

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Ear necrosis (EN) is a major welfare concern but there is poor understanding of the risk factors and causes. As stress is potentially implicated we investigated the effect of handling during lactation and mixing at weaning on EN development post-weaning. A total of 509 piglets were born to 37 sows over two replicates, and housed in farrowing crates. At farrowing, sows were blocked by parity and weight and their litters randomly assigned to either CONTROL (n = 238 piglets; 18 litters) or HANDLED (n = 271 piglets; 19 litters) treatments. Control piglets were handled 3 times during lactation: weighing at birth, processing (teeth and tail clipping) at ~5 days of age, and weighing 1d before weaning [~27 days of age]. Handled piglets were weighed an additional 5 times during the same period at intervals of 3 days. At 28 d of age, piglets were blocked by weight and sex and weaned into pens of 8-12 piglets. A 2×2 factorial design was used, so that half of the litters within each lactation treatment were weaned as INTACT litters (18 pens, n=206 piglets) and half were MIXED with piglets from ≥3 litters (18 pens, n=204 piglets). Pigs were individually assessed for fight lesions at 24h post-weaning (0-6, with score increasing with severity), and fight lesions on both ears were counted. Pigs were inspected for EN weekly from weaning until transfer to finisher accommodation (~75 d of age; 0-5, with score increasing with severity), then individually weighed at transfer. Data were analysed using SAS v9.4. There was no effect of either treatment on weaning or transfer weight ($P > 0.05$). Mixed pigs had higher fight lesion scores and a greater number of fight lesions on the ears ($P < 0.001$), but there was no effect of pre-weaning treatment on fight lesion scores ($P > 0.05$). There was no effect of pre-weaning or post-weaning treatment on EN severity ($P > 0.05$). The data suggests that under the conditions of this study, neither frequent handling nor mixing influenced growth or EN. Despite more aggression induced skin lesions in Mixed pigs, it seems that lesions caused by fighting are not risk factors for EN.

Identifying risk factors of piglet mortality at birth, before and after weaning

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Increasing piglet mortality represents a significant challenge for many pig farmers today. Stillbirth rates range from 5-10%, pre- and post-weaning mortality from 10-15%, and 1-4%, respectively, with some farms experiencing much higher rates. Effective measures to reduce piglet mortality rely on correctly identifying underlying causes and risk factors. While literature addresses various risk factors individually, understanding the intricate relationships among these factors is equally crucial. Therefore, a comprehensive literature review was conducted to summarize the main non-infectious and infectious risk factors related to piglet, sow and environment, along with their interconnections. Scientific articles were selected using Google Scholar, focusing on terms such as stillborn piglets, pre- and postweaning risk factors of piglet mortality, nest building and lying behavior of the sow. Key non-infectious sow factors influencing piglet mortality include breed, aggression, high piglet mortality in previous litter(s), post-partum dysgalactia syndrome, gestation and farrowing period, while infectious factors include PRRSV infections and fever-inducing diseases. Non-infectious piglet factors include poor fetal development, low birth weight, birth order, large within-litter variation, anatomical abnormalities and low weaning age/weight, whereas infectious factors involve diarrhea, meningitis, arthritis, skin disorders and systemic infections. Complex relationships exist within and between these factors. For instance, mortality rates of low birth weight piglets (< 1 kg) vary depending on birth order, ranging from 8% for the first 7 piglets born to 75% for subsequent ones. Whereas the combination of heavy piglets and young sows is more likely to result in farrowing complications and consequently higher mortality rates. With the goal of reducing piglet mortality, the PIGLIFE-project was launched. A comprehensive survey, based on this literature review, will be conducted on 60 farms to identify high-impact risk factors in a holistic manner. Furthermore, PLF tools (camera and RFID systems) will be developed to monitor farrowing, suckling behavior and feed intake in weaned piglets.

Session 23

Theatre 9

Environmentally enriched housing reduces disease severity in pigs after co-infection, with stronger effects when applied from birth than from weaning

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We investigated whether enrichment affects disease susceptibility of pigs studied in a co-infection model of Porcine Reproductive and Respiratory virus (PRRSV) and *Actinobacillus pleuropneumoniae* (APP). Pigs were either conventionally (CCH) or environmentally enriched housed (EEH) from birth onwards, or switched from CCH to EEH postweaning (CEH). At 60 days of age, 10 pigs per treatment were infected with PRRSV followed by APP infection on day 68. On day 71, lung morphology was studied to assess disease severity. Clinical manifestations and whole blood cell (WBC) counts and types, blood cell phenotypes and function of leucocytes were determined. Six additional pigs per treatment were euthanised before co-infection and bronchial-alveolar fluid (BALF) was tested. Results were analysed using mixed models. After weaning, EEH pigs had higher WBC and LPS treatment of whole blood cells revealed higher IL1 β and TNF α concentrations than CCH and CEH pigs. BALF of EEH pigs tended to contain relatively more macrophages than that of CCH and CEH pigs. In response to co-infection, EEH pigs had less exudative lung components of inflammation and the proportion of pigs with lung lesions tended to be lower than in CCH, with CEH in between. In conclusion, enriched housing reduced disease susceptibility to PRRSV-APP co-infection. In CEH pigs the response to the co-infection tended to decrease as compared to CCH pigs, but to a much lower extent as compared to EEH pigs. This indicates that enrichment from weaning is less effective in decreasing disease susceptibility than enrichment from birth.

Relationship between animal-based on-farm indicators and meat inspection data in pigs

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The aim was to validate slaughterhouse indicators (SI) collected during meat inspection as an alternative to on-farm animal welfare indicators. The assessments of 12 on-farm and 7 SI of 628 pigs from 3 different farms were combined into 3 indices, differentiated between on-farm and slaughterhouse: 1) limb health, 2) other organ health, and 3) respiratory health. Firstly, agreement parameters at animal level were calculated to ascertain whether the same welfare or health issues were identified on-farm and at slaughterhouse, taking the production period into account. Acceptable agreement was determined for the farrowing and fattening period, but not for the rearing period. A more detailed analysis shows a poor agreement 8 weeks and an acceptable agreement 4 weeks before slaughter. This indicated the SI pneumonia, pleuritis and pericarditis as possible estimators of fever and deviant behavior on-farm and the SI bursitis and joint inflammations as possible estimators of lameness. Secondly, the connection of slaughterhouse findings on the individual on-farm health indices was investigated; a significant influence of the farm on the limb and respiratory indices was determined. In general, some suitable SI were identified but a complete substitution of on-farm welfare assessment is not possible.

Session 23

Theatre 11

Identifying barriers and facilitators to implementing veterinary telemedicine in swine, poultry, and bovine production using a COM-B approach

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Efficient management plays a crucial role in reducing livestock diseases. Veterinary telemedicine, using communication technologies, offers innovative remote management solutions. Understanding farmers' and veterinarians' needs and constraints is vital for its future successful implementation. Therefore, four focus groups gathered the perspectives of farmers and veterinarians from both the bovine and monogastric sectors. Using the COM-B model of behaviour change, data was analyzed to identify barriers and enablers to the adoption of veterinary telemedicine. Farmers and veterinarians expressed consensus on two main benefits of telemedicine development: (1) the potential time saved travelling enabling quicker response to farmers' demands, not to mention the positive impact on the environment and (2) the desire for a holistic vision of health, including enhanced monitoring and analysis of health data. However, five major barriers remain: (1) disparities in the proficiency and availability of sensors and data across farms, (2) concerns about the loss of human interaction, (3) concerns about regulation of practices, (4) concerns regarding the value of remote services and (5) concerns about data and image hacking. Overall, both farmers and veterinarians demonstrate interest in how telemedicine could improve animal health and working habits, aligned with the goal of enhancing livestock health. However, they unanimously agree that barriers must be overcome to successfully implement veterinary telemedicine for livestock.

Implementation of sustainable parasite control strategies on extensively managed sheep farms

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Extensive farming systems play an important role in sheep production across Europe yet face many innate management and production challenges compounded by narrow economic margins which threaten their sustainability. Gastrointestinal nematodes pose a significant challenge to the productivity, health and welfare of grazing ruminants. While traditionally extensive sheep farms were not regarded as having significant parasite challenge, previous work has demonstrated a production limiting challenge and anthelmintic resistance, indicating the need for sustainable control strategies. However, current strategies are not optimised for extensive production systems. Optimised parasite control plans were developed and trialled for two years on nine extensive sheep farms. To measure their impact and practicality, production data and longitudinal parasitological data including faecal egg counts, anthelmintic efficacy testing and molecular species identification were collected. Semi-structured interviews were also conducted to capture management and socioeconomic-related impacts. Although solutions applied on farm were unique, they had similar core themes including increased evidence-based decision making, strategic use of anthelmintic and improved grazing management. Reduced anthelmintic use and improved lamb performance were observed, with farmers motivated to continue implementing these solutions beyond the project end. Implementation of sustainable strategies on extensive farms is thus achievable, however flexibility is required to fit individual management systems.

Feed intake, growth performance and gut health parameters of broiler chickens are positively affected by the supplementation of chestnut tannins in a dose-dependent manner

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Chestnut tannins may be a good alternative to the use of antibiotic growth promoters (AGP) thanks to their antioxidant and anti-inflammatory properties in broilers. To verify this hypothesis, 2,580 Cobb 500 broilers were subjected to 6 dietary treatments from 0 to 42 days: Uninfected Untreated Control (UUC), Infected Untreated Control (IUC), Infected Treated Control (ITC, with addition of salinomycin) and 3 treatments infected and supplemented with 3 levels of chestnut extract (KingBrown at 500g/T, 1kg/T or 2kg/T; named after KB0.5, KB1 and KB2). All broilers infected were challenged with a Necrotic Enteritis model. At 42d, individual live weights were significantly affected: UUC was significantly higher than IUC, ITC and KB2, while KB0.5 and KB1 were intermediate. For 0-42d FCR, ITC was significantly lower than IUC and UUC while KB0.5, KB1 and KB2 were intermediate. The ADG and ADFI were quadratically affected by KB supplementation with optimal concentrations between 0.5 and 1.0kg/T. The I See Inside (ISI) scoring, combining 9 gut health indicators confirmed the expected effects of the challenge on gut health with IUC having the worst score (9.2), then ITC (7.47) and UUC (6.32). Regarding the KB treatments, the lowest score was obtained with KB2 (5.55) while the other KB treatments were intermediate. In conclusion, low doses of KB supplementation tended to improve broiler performance under challenged conditions and could be an alternative to partially replace AGPs.

Use of infrared thermography for predicting the infectious etiology of subclinical bovine mastitis in a robotic milking system using machine learning

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This study assessed the potential of infrared thermography for predicting bacterial pathogens causing subclinical mastitis in pasture-raised cows using machine learning techniques in an automatic milking system. Eighty-nine lactating cows underwent thermographic analyses of their mammary glands over six months. Values (°C) corresponding to the mean, minimum, and maximum temperatures of the udders were recorded. Supervised machine learning techniques were used to predict mastitis and its infectious etiology using infrared thermography. Mixed binomial and multinomial logistic regression models were utilized. *Staphylococcus chromogenes* was the most frequently isolated microorganism (36.3%), followed by *S. aureus* (13%) and *Streptococcus dysgalactiae* (13%). The multinomial logistic regression technique showed high sensitivity (>0.95) in the identification of *S. aureus*, *S. uberis*, and *S. dysgalactiae* species. The AUC ROC for predicting the infectious etiology of subclinical bovine mastitis using infrared thermography was 0.90. Moreover, the SCC was significant in predicting mastitis caused by *S. aureus*, *S. uberis*, and *S. dysgalactiae* species. Infrared thermography demonstrated potential as a predictor of subclinical bovine mastitis due to *S. aureus*, *S. uberis*, and *S. dysgalactiae* in extensive cow rearing, a style traditionally seen as incompatible with robotic milking. The use of combined machine learning logistic models can optimize the identification of pathogens causing mastitis in bovine herds. Grant 2020/16240-4, São Paulo Research Foundation (FAPESP).

Prevalence and characterization of rib injuries in dairy cows: a neglected welfare issue of dairy production

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Overcrowded feeding areas, cramped stalls, inadequate cubicles, and slippery floors may lead to rib fractures in dairy cows. Despite the pain, these injuries are often overlooked by managers and technical staff. This study sheds light on this neglected injury to promote greater awareness within a context of animal welfare and the interaction of dairy holstein cattle with their environment. Data were collected in a dairy cattle farm, Gralha Azul Experimental Farm of the Pontifical Catholic University of Paraná – PUCPR, located at Fazenda Rio Grande, in the state of Paraná, southern Brazil. A description of the anatomic localization of the bone callus (affected side, rib, and position on the rib) was provided. A cohort of 72 cows, with an average age of 3.8 years and 2.7 lactations, underwent examination through palpation. Remarkably, 59.72% (n=43) of the cows exhibited rib fractures. Correlations were found for: age, regarding left medial fractures (R=0.306, P=0.007), and right ventral fractures (R=0.229, P=0.045); and lactations, regarding right (R=0.236, P=0.0395) and left (R=0.293, P=0.010) medial injuries. Additionally, a correlation was identified between right (R=0.245, P=0.032) and left (R=0.245, P=0.032) ventral injuries. Lesions were predominantly observed in cows with an average of 2.9 lactations. There was no correlation found between: dorsal and ventral fractures; nor dorsal and ventral injuries individually; nor age. Rib fractures, often unnoticed, impact cows' welfare over time. Older cows, more susceptible to fractures, face an increased risk of hipocalcemia and claw lesions, exacerbated by slippery floors, leading to falls and additional rib fractures.

Fertility and health traits of dairy cows on farms with short versus long productive lifespan profiles

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We compared fertility traits, health indicators and health management routines of Swiss dairy farms characterized by short versus long productive lifespans (SPL versus LPL). We analysed herdbook data from 142 Swiss dairy farms (n=142), farm questionnaire data (n=67), veterinary treatment data (n=64 farms) and data obtained during farm visits (n=30). LPL farms had a better fertility performance, i.e. lower number of inseminations per heifer, shorter average number of days open and shorter calving intervals. Number of antibiotic veterinary treatments due to fertility and other problems (i.e. others than fertility, udder or locomotion problems) were by tendency lower on LPL farms, while other veterinary medical treatments did not differ by productive lifespans. Although average somatic cell score and proportions of test day records with elevated somatic cell counts were significantly higher on LPL farms, udder health indicators remained within a healthy range and increases were not accompanied by higher treatment incidences for clinical mastitis. Moreover, the deterioration of udder health parameters was smaller than what could have been expected due to the age difference between contrasting lifespans. Locomotion scores and lameness incidence did not differ by productive lifespan profile. Cow health management routines did not significantly differ between farms of contrasting lifespan profiles, apart from a slightly higher proportion of LPL farms practicing abrupt drying off. We conclude that a longer productive lifespan is not at the expense of health, even if the SCC level increased with age. A focus on fertility, udder and claw health is crucial when aiming at a long productive lifespan.

The influence of sample size on fitting the true flock prevalence in animal welfare assessment impressively presented

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Data about the prevalence of health indicators give important information about animal welfare in a flock. Therefore, a number of protocols were developed. As a consequence of time needed for the assessments and the stress level for the animals, the sample size has to be limited. Animal welfare protocols like the Welfare Quality® Protocol, AWIN® or the KTBL® were developed, recommending the scoring of 50 up to 100 to 150 animals/flock. The aim of the present study was to systematically calculate sufficient high sample sizes for flocks of 50, 100, 200 up to 10,000 animals with acceptable margins of error of 1 to 10 % and confidence intervals of 90 to 99 %. Secondly, we simulated how the accuracy of the assessed prevalence changes when it deviates from the expected prevalence that was used for sample size calculation. In a last step, we additionally simulated the influence of detection bias due to observer or method errors. In the simulated data, an observation refers to a nonspecific but theoretically binominal trait. Analyses were conducted with JMP 15. Results presented in nice figures and tables show impressively the range of possible assessed prevalence for specific sample- and flock sizes and in how far the median differs significantly from the median of the true flock prevalence. We show e.g. that even in a small flock with 400 animals and an expected prevalence of 30% (CI= 95%) a sample size of 179 animals is needed what is more than the 50-150 recommended in the assessment schemes. Another example we present: an expected prevalence of 1%, in a population size of 400, leads to a sample size of 16. But what if the detected prevalence is 10%? The margin of error would be around 15%, what means the true prevalence could lay between >0 up to 25%. We think, the awareness of these ranges is important, not only for scientific investigations, but also for the practical case when reward or sanction systems are to be introduced on the basis of prevalence.

Introducing blood gas and ionogram analysis in swine farms for diagnostic and studies : sows reference values and practical aspects

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The acid-base balance in sow can be influenced by various factors, including feeding behavior, electrolyte balance, heat stress, and respiratory diseases. Disorders related to acid-base balance may be suspected in cases of well-being, performance and health problems such as lameness, sudden death, prolonged farrowing, and apathy. Additionally, nutritional and rearing strategies are evaluated in the field based on animal performance. Blood parameters could help evaluate the influence of certain controlled variables, and detect bias. Objectives: In the initial phase, our goal was to develop practical recommendations to ensure reliable results in terms of sampling and sample management. Then, the final objective was to determine, de novo, reference values for sows. Materials and Methods: We utilized the CLSI guidelines, adapted by the ASVCP. A direct sampling survey was conducted to establish de novo reference values for sows in early gestation, 5 days before and 5 days after farrowing. We included 12 French standard farms with indoor housing on concrete slatted floors and a diet consisting of commercial feed. These farms demonstrated high productivity results and sow mortality rates below 10%. Point-of-care analyzer was used on the farms. Results: The blood gas and ionogram analysis conducted on the farm yielded reliable results, provided that several pre-analytic requirements were met. A total of 440 healthy sow results were selected to determine the reference values. As a result, reference values have been established.

Session 23

Poster 19

Fecal biomarkers in piglets: assay validation

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Weaning is a stressful event that contributes to intestinal and immune system disorders in piglets, being an important problem in the swine industry worldwide. Measuring inflammatory biomarkers in fecal samples could be a good method to carry out a non-invasive diagnosis. The purpose of this study is to validate different analytical methods for assay biomarkers to predict intestinal inflammatory processes in pigs. Adenosine deaminase (ADA), alpha-1-antitrypsin (AAT), Calprotectin, immunoglobulin A (IgA), lactoferrin, lipocalin-2 (LCN-2), neopterin and myeloperoxidase (MPO) are potential biomarkers suitable for intestinal inflammatory disorders. Different kits and methods were evaluated following the recommendations of the American Society for Veterinary Clinical Pathology (ASVCP guidelines). Immunoturbidimetric assay for calprotectin, ELISA assays for LCN-2, IgA, neopterin and AAT and enzymatic colorimetric methods for ADA and MPO were adapted to fecal samples. Human specific kits were tested for calprotectin, lactoferrin, neopterin and ADA. Calprotectin, LCN-2, IgA, MPO and ADA achieved good accuracy obtaining an intra and inter-assay CV < 20%, recovery between 80-120% and $r^2 > 0,97$ for linearity under dilution. However, neopterin, lactoferrin and AAT did not reach satisfactory results due to low concentration samples and poor results in linearity study. Analytical validation is the first step in the development of new potential biomarkers of intestinal health. This will allow their use to monitor health status and welfare in weaning piglets.

Influence of pastoral management on the conservation of the cultural landscape and biodiversity of mountain pastures

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Mountain pastures are among the richest ecosystems in terms of plant and animal diversity. Moreover, they provide food, offer essential regulating ecosystem services to society (e.g., soil carbon storage, water purification, wildfire prevention) and are most valuable for tourism. Mountain agro-ecosystems are characterized by small-scale variability of abiotic factors (e.g., altitude, aspect, slope, bedrock). However, these systems are not natural, but the result of human pastoral management carried out for millennia. Traditional management has utilized the different feeding behavior of a variety of livestock species and breeds, as well as their specific impact on vegetation (e.g., trampling, nutrient and seed transport). Herd management techniques, animal selection and stocking rates, grazing periods and frequencies have been adapted to ensure sustainable use of the fodder resource. In recent days, traditional management is challenged by socio-economic and structural changes: direct payment policies influence management decisions, an increasing pressure of large predators complicate grazing, and climate change affects management conditions by altering pasture yield and its seasonal growth. In face of these changes, farmers have to newly define what site-adapted management means. Thereby, they have to avoid over- and underuse: in underused areas, wood succession leads to a loss of pastureland, biodiversity and appealing landscape. Management intensification, on the other hand, causes degradation, erosion, nutrient leaching and also decreasing biodiversity. However, new technologies, enhanced knowledge transfer and result-oriented policies have the potential to promote new site-adapted management strategies to maintain mountain pasture ecosystems.

Session 24

Theatre 2

Trends in the association of herbivores and pigs in the Massif Central

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The Massif Central in France is a mountainous and grassland territory, primarily dedicated to grazing livestock. On 15% of the national area, it concentrates 54% of the country's dairy sheep, 37% of the beef cattle, 33% of the meat sheep, 19% of the goats, 11% of the dairy cattle, 6% of the chickens and 5% of the pigs in 2020 according to the agricultural census. Out of 39,065 farms with a significant livestock (excluding farmyards) in 2020, 2,155 have granivores, mostly associated with herbivores. More precisely, 75% of pig farms have herbivores, beef cows in 62% of cases and milk cows in 24%. But these mixed farms have declined more sharply than the specialists over the last 10 years. This raises multiple concerns within the pig sector which has strong roots in this territory (quality signs, slaughterhouses, gastronomy, etc.). The APOR THE multi-partnership action-research program is interested in pig farms in the Massif central. A recent analysis carried out using individual data from the 2020 agricultural census, compared to 2010, showed that the decrease in the number of pig farms (-28% in 10 years) comes from a sharp decline in the pig-dairy association (-45%) and to a lesser extent in pig-beef cattle association (-25%). As a result, our drawn typology showed a large proportion of pig farms with beef cattle in 2020. Other results specify the extent of the changes in the herbivore-granivore association between 2010 and 2020. Our communication will enlighten the trends and the future of these mixed animal farms which remain essential for the agricultural and local economy of several sectors of the Massif and other mountain areas.

Contribution of livestock in organic agriculture development: a study on nitrogen flows in expansion scenarios in France

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The European Green Deal has established a goal of achieving a minimum of 25% of the EU's agricultural land under organic agriculture (OA) by 2030, compared to 9.9% currently. Nitrogen has been identified as the main limiting factor for the expansion of OA. Biological nitrogen fixation (BNF) by legume crops is the main nitrogen input available to OA supporting the entire OA agri-food system either through crop rotation or livestock integration. However, the contribution of organic and conventional livestock on the growth of OA, influenced by OA regulations, is barely addressed in the scientific literature. Here we examine the role of livestock in OA in the circularity of nitrogen of the agri-food system, in particular from grassland to cropland and its role on the proportion of animal proteins in human diets. We calibrated the ALPHA nitrogen budget model (Chatzimpiros and Harchaoui, 2023) based on national statistics and survey data to assess current nitrogen flow metabolism at the national scale for the case of France. We simulated various scenarios to achieve the 25% OA target, taking into account key variables including the proportion of ruminant and monogastric, feed composition and feed import, manure management efficiency and crop rotation strategies. In 2021, French OA demonstrated a 78% nitrogen self-sufficiency, with BNF contributing for 50% of nitrogen inputs. The remaining 22% of external inputs were evenly split between imported feed and conventional manure. The paper presents contrasting nitrogen circularity scenarios for OA that highlight the dynamics between organic and conventional livestock farming in terms of land use, self-sufficiency, food production and protein content in human diet.

The representations and attractiveness of livestock farming professions under the influence of societal issues. The French situation in a European perspective

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Livestock farming is the subject of a socio-technical controversy based on different levels of uncertainty. These relate to environmental and climate issues, animal condition, human and animal health, and, more generally, farming models and ethics. This controversy, which began in the 1980s, continues today. Even though livestock farmers have a rather good image, the general public, who know little about the realities of livestock farming, nevertheless have high expectations about livestock farming practices and models. Farmers, for their part, although aware of the challenges facing society, and sometimes proactive in taking these issues into account, often feel accused and questioned in their ways and even in their professional legitimacy. In this paper we will firstly present the current state of the societal controversy in France and Europe and propose 5 profiles of citizens described according to their expectations of livestock farming and their consumption of animal products. "We will secondly describe how farmers perceive this controversy, with those who share part of the criticism and are developing alternative models, those who feel that better communication should enable a compromise solution to be found, and those who feel under attack and react mainly negatively to questioning. By combining these representations with attitudes to change, we propose 4 livestock farmer profiles. We will thirdly present the effects of the controversy on the attractiveness of the profession, in particular its image in the general public and in the vocational universities, the conditions under which it is practised and the profiles of those who set up in business. In a fourth part we'll look at the various types of action taken by farmers and the industry: quality scheme initiatives, individual or collective communication campaigns, adaptation of farming practices and systems. Finally we will conclude with a discussion of the avenues to be explored to build a social consensus around livestock farming.

Life cycle and ecosystem services assessments provide opposite evaluations of the food and non-food contributions of livestock farming systems

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Life cycle assessment (LCA) and ecosystem services assessment (ESA) are often used for environmental evaluation. LCA is commonly applied to agro-ecosystems to assess the negative environmental impacts of food production (e.g. emissions of CO₂-eq.), whereas ESA is applied to assess the positive contributions of agro-ecosystems, such as the supply of regulating ecosystem services (e.g. pollination or erosion prevention). Here, we applied both frameworks to a selection of twelve contrasting French meat-production systems, including two ruminant species (sheep and cattle) and two monogastric species (chickens and pigs). The LCA indicated that ruminant systems had more negative impacts than monogastric systems, which is a well-known result. For example, the production of one kg of human edible protein emitted in average 280 kg CO₂-eq for ruminants, and 32 kg CO₂-eq for monogastrics. This result is due, in particular, to the methane emitted during the rumination process. Oppositely, ruminant systems can supply more regulating ecosystems services than monogastric systems. The mean respective scores related to these services were 2.42 and 1.15, respectively. This result is due to the presence of semi-natural habitats such as grasslands in ruminant systems. Our results show the opposition between the rating of food production by LCA, and the rating of non-food contributions by ESA. Our study stresses the need to reconcile these frameworks, as they can guide decision-making in opposite directions.

Session 24

Theatre 6

Beyond the Pigs: Socio-Economic Impacts of African Swine Fever (ASF) on Indigenous Breeds in Bulgaria and Eastern EU

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The study aimed to assess the impacts of ASF outbreaks on indigenous pig breeds, focusing on East Balkan swine (EBS), which represents Bulgaria's last local pig breed. Conducting extensive fieldwork, interviews with farmers and veterinarians (overall number=32), and a workshop with various EU stakeholders (>100 participants), the study explored the effects of ASF on EBS and backyard farmers in Bulgaria. The findings revealed the extensive destruction of backyard farms during the epidemic and a near-total loss of EBS herds. EBS farmers identified trade restrictions (77.8%) as the most significant consequence; 66.7% of EBS farmers reported direct impacts, but only 60% of affected farms received subsidies, often insufficient to cover their losses (60% of their income came from meat selling that is still forbidden for EBS farmers). ASF was confirmed as a major challenge for other local breeds like Bazna Pig (RO) and Mangalitz (HU), even if significant disparities in financial support and implementation of preventive measures emerged when comparing different EU realities. The study emphasized ASF's long-term risks, including disruptions to local economies, loss of cultural heritage, biodiversity reduction, and increased land abandonment. Comparisons among European realities revealed significant disparities in measures, such as financial support to local breeds. There is a pressing need for strengthened collaboration among veterinary authorities and rural development stakeholders, aligning with EU green policies during sanitary crises. Tailored policies and regulatory adjustments are essential to preserve animal biodiversity, traditional breeding farms, and linked economies. Finally, there are currently no clear indicators to quantify the economic, social, and cultural value of EU indigenous breeds, reflecting the multifaceted roles of livestock.

Biopolymers of animal origin: production, properties and applications

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In the pursuit of sustainable and biocompatible materials for pharmaceutical, biomedical and cosmetic applications, biopolymers derived from animal sources have emerged as promising candidates. These biopolymers offer unique properties, including biodegradability, biocompatibility, and low toxicity, making them valuable in various formulations. Gelatin, collagen, chitosan, and hyaluronic acid are among the most important industrially relevant biopolymers derived from by-products of animal industry. These biopolymers have diverse range of pharmaceutical, biomedical and cosmetic applications. For example, gelatin is an important pharmaceutical material used for manufacturing of hard and soft capsules as well as suppositories. Collagen is commonly used as a dressing in wound management as well as a matrix for tissue engineering. Chitosan-based formulations exhibit mucoadhesive properties, enabling targeted drug delivery to mucosal surfaces such as the gastrointestinal tract, the eyes and nasal cavity. Hyaluronic acid-based products offer visco-supplementation in osteoarthritis treatment, lubrication in ophthalmic solutions, and hydration in dermal fillers. Some animal-derived biopolymers such as heparin and insulin have specific pharmacological activities. Heparin is used as a blood anticoagulant and insulin is a hormone that is used in the management of diabetes mellitus. In some cases, these materials could be chemically modified further to impart additional properties, for example, adhesive properties of gelatin could be significantly improved by introducing unsaturated functional groups through reaction with crotonic, itaconic, and methacrylic anhydrides. Methacrylated gelatin is also commercially available as a material for producing hydrogels for tissue engineering. Mucoadhesive properties of chitosan could be altered by grafting short-chained non-ionic polymers; these derivatives could be used to formulate mucus-penetrating nanoparticles for drug delivery. This talk will provide an overview of animal-derived biopolymers, describe some of the methods used for manufacturing these materials, highlight their applications in pharmaceuticals, biomedical, and cosmetic industries, as well as discuss the challenges associated with the use of materials of animal origin.

Session 24

Theatre 8

Valorization of a waste from food industry: bovine hide to leather

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From cattle's slaughtering process, great amounts of hides are obtained as by-product. Excluding the small portion used by edible gelatine industries, the majority of that undergoes tanning operations resulting in genuine leather, nowadays in an increasingly sustainable way (1). Raw hides not intended to become leather or edible gelatine must be destroyed. Taking into account the ratio between the weights of raw hide and whole animal, this fraction is very low, while considering the economic value of leather compared to that of cattle's food derivatives, leather is high value-added final manufacture product. Hide's tanning is one of the first examples of circular economy. A waste is used as a raw material for another industrial sector to create valuable products, placed on several markets, e.g. shoes, leather goods, garments, upholstery, car interiors. Recently, a number of leather substitutes emerged, some of them claimed as "sustainable-vegan leather". However studies show that, at the moment, genuine leather is far superior on the basis of different aspects:– Renewable carbon percentage in leather, as the starting point is livestock, is on average higher than alternatives (2)– Most of the alternatives is made by synthetic polymers and fibers, those manufacture starts from fossil resources (3)– Taking into account the technological and mechanical properties, leather surpasses several substitute materials (3): leather goods last longerReferences:1) Kanagaraj, J., Panda, R. C., & Kumar, V. (2020). Trends and advancements in sustainable leather processing: Future directions and challenges—A review. *Journal of Environmental Chemical Engineering*, 8(5), 1043792. Carcione, F., Defeo, G. A., Galli, I., Bartalini, S., & Mazzotti, D. (2023). Material Circularity: A Novel Method for Biobased Carbon Quantification of Leather, Artificial Leather, and Trendy Alternatives. *Coatings*, 13(5), 8923. Meyer, M., Dietrich, S., Schulz, H., & Mondschein, A. (2021). Comparison of the technical performance of leather, artificial leather, and trendy alternatives. *Coatings*, 11(2), 226

Cross-analysis of crop-livestock integration, sustainability and biodiversity on 10 French experimental farms
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Over the past 60 years, crop rotations have been simplified and now these systems have environmental, social and economic limitations. Putting crop-livestock interactions back at the heart of food production systems is a possible avenue for improvement. This study tried to evaluate the link between the level of crop-livestock interaction (CLIL) and farm performances in terms of biodiversity and, more generally, sustainability. The IDEA4 (Farm Sustainability Indicator, version 4) and BIOTEX (Biodiversity for the Farm) methods associated with NiCC'El (Crop-Livestock Interaction Level) enable this exploration. These methods have been implemented on 10 french experimental farms and scores were analyzed using Kruskal-Wallis tests. Of the 10 farms, 4 are classified as low CLIL, 3 as intermediate CLIL and 3 as high CLIL. As the farms do not have profitability as their primary objective, the economic dimension is often the limiting factor in IDEA4. The CLIL is significantly correlated with the score for the socio-territorial dimension, and a positive correlation trend is observed with the agroecological dimension. Quality and diversity of products sold, feed and fertilization autonomy, sobriety in the use of resources and reduction of impacts on ecosystems are thus more favorable in farms that have high CLIL. In BIOTEX evaluations, high CLIL farms fare better than low CLIL farms in terms of crop and grassland management, but seem to contribute less to land-use diversity. All these tools complement each other, but the time required to collect the data needed to carry out these diagnoses is a limiting factor when considering extending this study to a large number of commercial farms

Session 24

Theatre 10

Bee friendly livestock systems

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The introduction of legumes and brassicas into grasslands increases floral resources that benefit pollinators, native wildlife, and a range of ecosystem services with positive economic consequences. The objective of a couple of studies was to assess bee species presence in a multi-species forage trial and a grazing trial at North Florida. Thirty plots of 1.8 x 3 m with 0.9 of allies between them. Emergence traps were set over the 30 plots during the last week of April to June for two years. Every week the traps were moved inside the same plot and the insects collected were placed in vials with 70% alcohol for posterior identification. The flower counting and the soil cover percentage were performed every two weeks from April to June, and once per month, a 5 min pollinators visual surveys were performed. In the grazing trial, three treatments—grass with N fertilizer, no fertilizer, and a mixture of grasses with legumes—were each replicated three times. Bowl traps were deployed for 24 hours once per month over 2 years. In the multispecies trial bee abundance and richness were notably higher in the grass-legume system. Visual survey results indicated a significant effect of the year: in the first year, there was a statistically significant difference between treatments and the control ($P < 0.05$), as well as among the number of functional groups. However, in the second year, treatments did not significantly differ from the control. In the grazing trial, the abundance of bees was notably higher in the grass-legume system compared to the Grass+N system ($P = 0.01$). Additionally, a significantly greater density of flowers was observed in the grass-legume system compared to Grass+N ($P < 0.05$). In conclusion, these studies demonstrate that well-managed grasslands play a crucial role in enhancing landscape structure and flower resources, thereby offering stability to bee communities.

Session 24

Theatre 11

Closing remarks

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Since 2013, the ATF-EAAP Special Session during the EAAP Annual Meeting aims to bring together animal science with practice of animal production and connect researchers, policy makers, industry representatives and societal organisations. Every year, a different topic is addressed during this half-day session. Once again, and for the 4th time, the EAAP Livestock Farming Systems Study Commission and the ATF Special Session have joined into a one-day symposium. The theme is Livestock are more than food. The discussion will continue in November in Bussels during the ATF symposium.

Session 25

Theatre 1

Formulating balanced diets with local agricultural by-products from Greece as sustainable feed ingredients of *Tenebrio molitor* larvae

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Agricultural by-products hold many advantages as potential insect feeding substrates. Taking into account the growing demand for insects as alternative proteins, through the valorization of by-products as insect feedstocks, waste management is promoted. However, to meet the nutritional requirements of farmed insects, composed diets combining diverse ingredients should be designed. In this context, the present study evaluates local agricultural by-products from Greece stemming from the production of triticale, oat, barley, cotton cake, lupin, hempseed, and rice as diet ingredients for *Tenebrio molitor* larvae. Briefly, four distinct groups of isoproteic and isolipidic diets were formulated, each group varying in protein content from 17 to 25% and lipid content from 4.1 to 4.8%. Within each group, different diets were designed, incorporating the aforementioned by-products at various percentages. Through this approach, we aimed to explore the synergistic effects of different agricultural residues on insect growth and development. In a first trial, the growth and performance of 50 newly-hatched larvae fed with the different diets was evaluated in lab scale. In a second trial, the diets that performed best were evaluated in pilot scale with 10,000 larvae. The results showed that diets with balanced ingredients deriving from local agricultural by-products are suitable as feeding substrates for *T. molitor* promoting larval growth and sustainable insect farming. This research is supported by the EU-PRIMA program project ADVAGROMED (Prima 2021 – Section 2).

Yellow mealworm larvae fed on Italian by-products

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To improve sustainability in yellow mealworm rearing, the utilization of by-products needs enhancement. For this purpose, various by-products were collected: wheat groats, wafer dough, dry distillery stillage, coffee silvery film, rice by-products, Christmas dessert, breeding and feed waste. These by-products were used to formulate 3 iso-proteic, -lipidic, and -energetic diets (TM1, TM2 and TM3), and wheat bran was tested as a control (C). The trial was conducted on a large scale (10000 larvae/replicate and 4 replicates/treatment) starting from 4 weeks old (WO) larvae. The sampling operations was performed every week until the difference in the percentage of larval weight between two consecutive sampling was lower than 50%. At the end of the trial, the number of larvae was estimated, and the biomass was weighed to calculate survival rate and feed conversion ratio (FCR). A sample of larvae was collected for chemical analyses (dry matter, DM; crude protein, CP; ether extract; ash and chitin). Data were analysed by One-Way ANOVA (SPSS, V20.0.0). No differences in terms of weight were observed from 5 to 7 WO larvae, while the C, TM2 and TM3 larvae reached higher weight than TM1 at 8 and 9 WO ($p < 0.01$). Dietary treatment did not influence the survival rate and FCR. Larvae fed on C diet showed the lowest DM and the highest CP ($p < 0.01$). Both C and TM3 larvae showed a larger amount of ash than the other diets ($p < 0.01$). The TM1 and TM2 larvae were fatter than C and TM3 groups ($p < 0.001$), which were statistically different ($p < 0.001$). No difference in terms of chitin was detected. In short, the TM3 diet, characterized by the inclusion of rice by-products, demonstrated greatest performance in both growth and chemical composition.

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Theatre 3

WP2 – Local agricultural by-products as substrate for insects: results in Portugal

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The ADVAGROMED WP2 aims to valorize local agricultural by-products that have no economic value for small farms and agri-food industries in Italy, Greece, Portugal, Spain, and Morocco, which could be exploited as insect feeding substrates. Four by-products were selected for insect diet trials based on their availability throughout the year: white melon, broccoli, olive pomace, and wheat bran. Diets were designed using linear model software to obtain different by-product mixtures with a relative moisture level of 70% and a crude protein content (~5%) comparable to a control diet composed of Gainesville compound feed. Juvenile Black Soldier Fly (BSF, *Hermetia illucens*) larvae were inoculated in boxes (40×60 cm) in three replicates per diet and allocated in the environmentally controlled room. The bioconversion period was monitored daily, and the environmental conditions were adjusted accordingly over time. The following parameters were determined for each diet: total larval weight (fresh), individual larval weight, feed conversion ratio (FCR), and survival rate. Based on performance results (1.8 kg/box of larvae, 78.98 mg per larva, 77% survival rate, 7.53% FCR), diets composed of melons and olive pomace were selected to feed the BSF larvae, aiming to produce fresh larvae (and frass) for use in WP3 by the SERIDA partner. Moreover, surveys are being conducted to outline the reality in Portugal regarding the availability and utilization of by-products and other potential insect rearing substrates. The objective is also to design strategies for the storage of these by-products to ensure their availability for insect production, thereby laying the groundwork for the growth of a new bioindustry in Portugal.

Effect of different diets on the larvae growth performance of Black Soldier Fly « *Hermetia illucens* »

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In the search for new sources of ecologically sustainable feed, numerous research studies have demonstrated the alternative role that the larvae of the black soldier fly *Hermetia illucens* could play in animal nutrition. Our study was carried out with the aim of researching potential diets of high nutritional value of agricultural by-products collected at Ain Taoujdate, and testing their effects on the growth performance of larvae. The selected by-products were apple, pumpkin, beetroot, eggplant and gainsville. The by-products, were prepared at the required moisture (70%). The trial consists of grinded by-products of 5 diets. The juvenile larvae were incubated in a growing room (26°C and 70% of humidity). The daily weight, length, width, survival, total amount and dry matter of larvae was registered. Our results showed that larvae are growing fast using gainsville, eggplant and moderately with beetroot. These three diets showed high values in terms of survival rate and dry matter of larvae.

Session 25

Theatre 5

Effect of agro-industrial by-products-based diet on the gene expression of antimicrobial peptides in *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) larvae

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Several researches confirm that the exploitation of agricultural by-products for insect rearing could offer a possibility for considerable reduction of the production cost, whereas it contributes to the enhancement of the sustainability profile of insect production. Potential rearing substrates are often evaluated taking into account their influence on larval survival, growing performance and substrate reduction capacity. However, variations in diet composition could also influence insect immune responses and therefore their susceptibility to entomopathogens. In the frame of the ADVAGROMED project, the effect of the diet on the gene expression of antimicrobial peptides (AMPs) was evaluated in *Tenebrio molitor* L. (Coleoptera: Tenebrionidae). Analyses were performed on mature larvae reared on wheat bran, used as a control diet, and on three agro-industrial by-products-based diet containing: i) wheat groats, wafer dough, vinasse, coffee silvery film, breading waste and feed waste; ii) wafer dough, breading waste, feed waste and panettone; iii) rice husk, rice chaff, rice middlings, breading waste and feed waste. AMPs encoding gene expression levels were evaluated through RNA extraction and RT-qPCR analyses. The gene expression of three AMPs (one Cecropin, one Coleopterecin and one Tenecin) was assessed. The influence of diet on the expression of different AMPs was observed in our trials highlighting the importance of considering this aspect during diet formulation in large-scale productions. Diets that are not favourable to the immune system could be supplemented with products that act positively on the insect immune responses (supported by ADVAGROMED PRIMA project).

WP3 – The use of live insect larvae as feed ingredients for local poultry strains

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The WP3 of the ADVAGROMED project aims to investigate animal performance, welfare and health, and product quality of local poultry breeds reared in four Mediterranean countries (Italy, Greece, Spain and Morocco) and fed live insect larvae as feed ingredients of diets formulated using local raw materials. *Hermetia illucens* (HI) and *Tenebrio molitor* (TM) live larvae will be included at 5 and 10% (on dry matter) of the daily feed intake of the birds, by partially or completely replacing a feed mix (characterized by high-quality, highly digestible, and less sustainable ingredients) that will mimic (being isonitrogenous and isonergetic and containing similar amino acid contents) their nutritional profile. Two egg-type (Bionda Piemontese [Italy, fed TM live larvae] and Pita Pinta [Spain, fed HI live larvae]) and meat-type (Black-Bronze turkey [Greece, fed TM live larvae] and Beldi chicken [Morocco, fed HI live larvae]) strains will be reared (6 replicates/diets, 10 birds/diet) for 12 weeks (egg-type strains) or 60 days (meat-type strains), and bird productive performance (growth indices, egg production rates, and carcass traits) will be recorded. Animal welfare (by means of plumage status, behaviour, and corticosterone analyses) and health (in terms of blood and postmortem investigations) will also be considered, along with physico-chemical quality and consumer acceptance of the final products (eggs and meat).

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Theatre 7

Responses of Pita pinta laying hens fed live black soldier fly (*Hermetia illucens*) larvae in Northern Spain

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The use of insects as bioconverters of local by-products to feed laying hens could be an efficient alternative to reduce the use of imported feedstuffs and their associated environmental impact. However, little is known about their potential for poultry production systems based on autochthonous breeds. Within the ADVAGROMED project, the animal performance, welfare, health, and product quality of Pita pinta laying hens fed live *Hermetia illucens* (HI) larvae were studied. Three treatments (control, H5 and H10) with 3 replicates and 10 birds/replicate were applied for 12-weeks. HI live larvae were added at 0% (control), 5% (H5) and 10% (H10) of the expected bird daily feed intake, by partially or completely replacing it by a feed mix that mimicked their nutritional profile (but with less sustainable ingredients). Egg production and physico-chemical quality, animal welfare (plumage status and corticosterone levels), and health status (blood and faecal parameters) were assessed in three periods (T0, T1 and T2). Preliminary analyses showed no differences in life weight of Pita pinta hens, egg shape index and eggshell thickness between treatments. Egg shell weight was lower in H10 eggs than in the other treatments in T1. The highest values of Haugh Units and yolk index were found in control and H10 eggs respectively during T1. This study provides important information about the potential role of live larvae as feed ingredients for sustainable local poultry productions.

Insect frass and poultry manure: a novel circular economy approach for sustainable vegetable production in the Mediterranean regions. The PRIMA project ADVAGROMED experiences

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The goals of the ADVAGROMED is to integrate organic fertilizers such as insect frass (IF) and poultry manure (PM) into sustainable agricultural practices to enhance local agro-biodiversity. The project aims to achieve several objectives, including testing insect-derived products as biofertilizers, evaluating their effects on plant growth and soil fertility, and developing guidelines for their use in Mediterranean agriculture. Agronomic trials have been conducted in growth chambers and open fields by various project partners distributed in different Mediterranean countries. We tested the impact of IF and PM on the crops performance and nutritional characteristics of local varieties (Brassicaceae, Solanaceae, Asteraceae, Fabaceae, Poaceae, Amaryllidaceae). The tests conducted in growth chambers and in vitro revealed a potential phytotoxic effect of both IF and PM at high doses. The results from field trials indicate that the use of IF and PM allows for yields similar to those obtained with chemical fertilizers, without compromising the quality of tomatoes (°Brix, titratable acidity, and mineral content). In addition, an increase in various productivity indicators has been observed in peppers. Supported by PRIMA 2021 ADVAGROMED project.

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Theatre 9

The effect of added insect frass as a soil amendment in the NPK content of two pepper varieties

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Insect frass is a waste material high in organic C and bioavailable macronutrients, like N, P and K. In a 3-month pot experiment, we tested frass from *Tenebrio molitor* larvae and poultry manure as soil amendments in the cultivation of two pepper plant varieties, “Bachovitiki” and “Florinis”. Frass was added at 0.5 and 1%, and poultry manure (PM) at 2.5 and 5% to two different soils (one of light texture-Larissa, and one of medium texture-Velestino). There were also two control treatments, one negative (NC; no additions) and one positive control (PC; inorganic fertilizer). This experimental design resulted in 240 pots (6 treatments × 2 soils × 2 pepper varieties × 10 replicates) of a capacity of 10 L each. We found that leaf N content increased significantly in both soils and in both pepper varieties with added frass and PM. Irrespective of the soil, N at NC was ca. 2% and it increased significantly in the highest PM rate of 5% to the levels of 3.3% in Bachovitiki and 2.6% in Florinis (in the sandy Larissa soil); similar was the case in the loamy Velestino soil. This shows that the rate of increase of N in pepper leaves was variety-dependent rather than soil-dependent (i.e., there were no considerable differences among the two soils when same frass and PM treatments were compared). As for P, it was 1.5‰ at NC in the Larissa soil in both varieties, while it was 2.5‰ in the more fertile Velestino soil. Although P uptake was soil-dependent, with added frass and PM the P levels increased significantly in both soils and in both varieties, reaching 3‰ for both varieties in Larissa and 4‰ (Bachovitiki) and 4.5‰ (Florinis) in Velestino. Similar were the trends for K: at NC pepper leaves contained K in the range of 2.5-3% in Larissa and 3-3.5% in Velestino, indicating the sufficient levels of available K in both soils even at NC, probably due to the over-fertilization of these agricultural soils. We conclude that both frass and PM had a positive effect in the nutrition of the two tested pepper varieties in the two soils with varying texture. This research is supported by the EU-PRIMA program project ADVAGROMED (Prima 2021 – Section 2).

Hermetia illucens frass and Pita pinta laying hens manure as biofertilizers for Asturian crops: effects on seed germination and growth

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The demands for crops, goods and services boost the use of fertilizers with adverse environmental impacts. Alternative local fertilizers are essential to reduce costs and improve soil health. The ADVAGROMED project explores the effects of frass (F) from *Hermetia illucens* rearing and manure (M) from Pita pinta hens fed with live *H. illucens* larvae on seed germination and seedling growth of *Brassica oleracea* L. var *Viridis*, *Lactuca sativa* L., and *Phaseolus vulgaris* L. var *Verdina* in Asturias, Spain. Four biofertilizers were tested: F and 3 types of M from hens fed with 0%, 5% and 10% larvae addition to their diet, respectively. Ten liquid extracts, from 10% to 100% concentration were prepared from each biofertilizer (pre-treated at 70°C/1 hour) with distilled water (ratio 1/10, w/v) and filtration pore size of 0.8µm. Three replicate petri dishes per treatment were sown with 25 or 15 seeds for *L. sativa*/*B. oleracea* and *P. vulgaris*, and kept at 24°C, 70% RH, 12-hour photoperiod for 7 days. The number of germinated seeds were recorded daily whereas radicle and hypocotyl length were measured after 7 days. Germination index, germination percentage, coefficient of velocity of germination, seed vigor index, mean germination time and T50 were calculated. The four biofertilizers up to 30% concentration improved all germination indexes without surpassing tolerance limits for all species. F and M met nitrogen demands, suggesting they are viable organic alternatives for these crops.

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Theatre 11

Effect of insect frass on the growth of barley crop

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Eugénio 140, 2005-079 Várzea, Portugal

In the context of current climate change, farming methods exploring to decrease their environmental impact are increasingly sought after in recent years. Insect frass was recently used as a sustainable biofertilizer with multiple benefits for crop cultivation. Our investigation aimed to assess the effect of insect frass of black soldier fly on the growth of barley crop in Morocco. Field trials were carried out in Ain Taoujdate, where physicochemical analysis of the soil was released. Based on N content in the insect frass and the rate of N used as reference for barley crop, 6 treatments were used: insect frass at the rate of 1.42, 2.85, 5.7, 11.4 t/ha, negative and positive controls. Morphologic, agronomic and physiologic parameters were measured, including seed germination, plant growth, yield, plant height, flowering dates, dry matter of roots and shoots, number of leaves, leaf area, content in N/P/K and chlorophyll). The aim of our study is to demonstrate the useful rate of application of insect frass for the optimal barley growth under Moroccan climate conditions.

Insect Frass Fertilizer: A Green Solution for Tomato and Brassica Microgreens

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In the near future, a significant increase in insect production is expected, leading to substantial quantities of frass-based residues. Within the framework of the circular economy and agricultural sustainability, these residues can serve as organic fertilizers in agricultural settings, substituting mineral fertilizers, or find utility in controlled environments like greenhouses and vertical farms, where they can be used as cultivation substrates and rich sources of nutrients. This research aimed to assess the impact of frass from *Hermetia illucens* (H) and *Tenebrio molitor* (T) on the nutritional composition of tomato, mizuna and rapini microgreens, in open field (tomato) and growth chamber (microgreens). In both vegetable productions, the utilization of frass exhibited no discernible effects on yield or visual quality. In tomato, the application of T frass didn't induce variations in nutritional parameters in comparison with those observed for the control samples (color, °Brix, titratable acidity, mineral elements). In mizuna cultivated with H frass, there was an increase in mineral content such as Fe (28%) and Zn (147%) compared to the control peat, thereby offering potential health benefits for consumers. Moreover, no contamination by *Salmonella* spp. and *Escherchia coli* or heavy metals was detected in the microgreens. Preliminary results here reported suggest the possible application of frass in different horticultural production in both open field and greenhouse environments. Supported by PRIMA2021 Advagromed project.

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Theatre 13

Metagenomic insights into the effect of frass fertilizer application on rhizosphere microbial community: the case study of Italian turnip

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The intensification of current agricultural practices and production systems is a primary cause imputable for the deterioration of soil quality, erosion, organic matter depletion and decline of soil biodiversity. Organic fertilization is proven to be useful in preserving and enhancing soil microbial communities. Considering the rising of insect farming in the near future, integrating insect frass, as organic fertilizer, into sustainable agricultural practices could enhance and preserve local agro-biodiversity. This research aimed to evaluate the impact of frass, derived from *Tenebrio molitor*, and poultry manure fertilizer application on rhizosphere microbial community of *Brassica rapa* local variety through metagenomics. An open field experiment was set from October 2023 to February 2024 using different fertilizers: chemical fertilizer, organic fertilizer, *Tenebrio molitor* frass, poultry manure and their mixtures. Metagenomic analysis was conducted on rhizosphere samples for each treatment and compared with uncultivated soil sample, in order to evaluate microbial changes during the cultivation period. Statistical comparison among taxonomic categories was performed using STAMP software. Differences between groups were analyzed using Welch's t-test ($p < 0.05$), and the Benjamini–Hochberg false discovery rate (FDR) method ($q < 0.05$) was used. Results indicate that the application of insect frass poultry and manure fertilizer is able to modulate the rhizosphere microbial community. Supported by PRIMA2021 Advagromed project.

Social LCA of a small-scale insect-fed broiler farm

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Application of social LCA (SLCA) is challenging due to the lack of standardization. This research aims to contribute to standardization of SLCA in small-scale farming of black soldier fly larvae (BSFL) farm, and small-scale broiler farm which supplements the broiler feed with live BSFL. For both the insect farm and broiler farm 2 scenarios were considered (with predominantly manual production, and predominantly automated) with 2 additional scenarios for the broiler farm (with and without BSFL supplementation). The study followed the cradle-to-farm gate approach. The grading of social risks was done by assigning a grade between 1 (committed) and 5 (not assessed – unreliable sources) to each of the previously defined 8 categories. Following this methodology, it was observed that the introduction of insects did not have a significant impact on SLCA, increasing the results by 0.12 and 0.13 points for manual and automated broiler rearing, respectively. On the other hand, a clear positive impact of automation was observed, leading to a difference of 1.07 points in the case of BSFL rearing, 0.72 points in the case of BSFL-supplemented broilers, and 0.73 in the case of control broilers. The decisive categories were Health & Safety (which included the risk of allergenicity of handling insects), Working hours and Equal opportunities/Discrimination. It can be concluded that, while BSFL inclusion did not lead to a decisive change, an improvement of the social sustainability of insect and broiler farming can be achieved through an increase in the level of automation.

Session 25

Poster 15

ADVanced AGROecological approaches based on the integration of insect farming with local field practices in MEDiterranean countries. The PRIMA project ADVAGROMED

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In intensive agricultural and animal production systems, the primary contributors to biodiversity loss are the conversion of natural habitats into agricultural land and the extensive application of chemical inputs (fertilizers and pesticides). The incorporation of agroecological methods into existing agricultural systems presents a sustainable approach to safeguard and enrich endangered farming biodiversity while simultaneously boosting ecosystem services. To enhance the resilience of agro livelihood systems, ADVAGROMED seeks to create an innovative and comprehensive food system based in agro-ecological principles and circular economy practices. By-products from local agricultural processes are used to grow insects, resulting in two distinct products: 1) insect frass, serving as a bio-product to enhance soil fertility, provide plant protection benefits, and boost soil microbial biodiversity, thereby reducing the reliance on mineral fertilizers and chemical pesticides; and 2) live larvae, used as feed for local poultry breeds to optimize animal performance, health, and product quality, consequently decreasing dependence on imported feeds.

Local agri-food wastes and by-products in Asturias (NW Spain): current situation and potential role as substrates for insect rearing

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The agri-food sector in Asturias (NW Spain) could generate by-products of interest for insect rearing, although their suitability depends on diverse factors. The objective of this work linked to the ADVAGROMED project was to identify, select, quantify and characterize a portfolio of these local by-product sources for insect feed. The identification was based on information gathered from official databases and reports of previous related projects, followed by a review of the main regional agrarian productions, their location and socioeconomic context. Additional wastes (i.e. from gardening) or marine algae (fresh or by-products from its processing) were added to the preliminary inventory. The selection was based on: economic or cultural importance of the productive sector, quantity, location, availability over time, accessibility and homogeneity. A total of 24 by-products and other potential sources for insect feed were selected from the following productive sectors: kiwi, blueberry, horticulture production, bean, cereal, cider, animal feed, algae and gardening. Subsequent surveys to the most relevant agri-food companies/producers were performed to quantify and contextualize these sources. Data about parameters such as their geographical distribution, quantities, availability, current usage and storage/treatment strategies were collected. Moreover, the nutritional composition (moisture, crude protein, lipids, crude fiber and ashes) of each source was assessed to complete its characterization. Algae and cider residues/by-products may be of special interest due to their quantity, location, and availability throughout the year.

Session 26

Theatre 1

The state of camel genetics: recent phenotyping approaches and genetic findings

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Dromedary camels are domestic animals of exceptional adaptive traits, functional and production attributes, and aesthetic phenotypes. Nonetheless, these phenotypes, especially heritable ones, are poorly documented and rarely characterized for genetic studies. Despite the recent development of genomic resources for dromedaries such as reference genome and SNP arrays, the limiting factor for fruitful genetic studies remains “the phenotype”. Therefore, the objective of this talk is to (1) provide a modern image-based phenotyping approach for dromedaries to document and describe categorical and continuous traits, and (2) report recent genetic findings on discrete aesthetic traits within the show-camels of the Arabian Peninsula, ‘Mezayen’ camels. We developed mobile-phone application, *Samplease*, to systematically collect detailed sample information, pedigree records, and photographs. Using over 500 photographs of collected camel samples and using a geometric morphometrics framework, we characterized categorical traits including: coat colors, hair lengths, crimp-types, ear and mouth attributes, nasal bridge curvatures, and overall head shapes. We further utilized the images to perform image-based geometric morphometrics to investigate body shape and size variation (e.g., hump shape and position) as means to phenotype external discontinuous traits. These phenotyping efforts enabled the identification of two variants within *FGF5* in relation to hair length variation and fiber quality. Furthermore, genome-wide association studies were conducted on the characterized phenotypes, particularly the categorical ones, and phenotype-associated regions were identified and awaits molecular analysis and validation. Our developed tools and adopted phenotyping approach can readily be applied to other camel populations to unravel possibly more and different phenotypic variations and enable more genetic discoveries.

The relevance of camel research and development for One Health

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One Health is an integrated, unifying approach that aims to sustainably balance and optimize the health of people, animals and ecosystems.” (www.who.int). For biodiversity conservation and public health newly emerging zoonotic disease outbreaks pose serious risks. Pathogens are omnipresent and by definition detrimental to their hosts, thus exerting high selection, which can result in reciprocal adaptation between the antagonists, an evolutionary arms race. Metagenomic next generation sequencing (mNGS) is a pathogen-agnostic detection method, and in combination with open-source platforms well suited to identify pathogens from genomic data. mNGS is a powerful tool to simultaneously retrieve One Health genomic information of (i) the host including its immune genes and microbiome, and (ii) pathogens present in the collected samples. In my presentation I will give examples of host immune response to emerging disease, e. g., dromedaries to Middle-East Respiratory Syndrome Corona Virus (MERS – CoV) infection. I will also give a preview about our current pathogen detection by screening Illumina short reads. Finally, I will show the importance of camels in the One Health context.

Session 26

Theatre 3

Recent discoveries in two milk proteins genes (CSN1S2 and LALBA) revealed useful markers for gene's assisted selection in camels

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Milk proteins are widely recognized for their high quality and digestibility. In this study, the coding genes of two major milk proteins, α s2-casein encoded by the CSN1S2 gene, and α -lactalbumin encoded by the LALBA gene were characterized in Old and New World camelids. Promoters and coding regions of these genes were sequenced and analyzed using molecular techniques and bioinformatics. Notably, a mutation was identified in the 3'-untranslated region (UTR) of the CSN1S2 in dromedary camels, at position g.15110G>T. This mutation was found to affect microRNA binding sites with a potential impact on miRNA 4662-3p, involved in mammary function. Furthermore, a novel mutation g.3639C>G was identified in the exonic region of CSN1S2 in Bactrian camels, resulting in an amino acid variant at position 36 (p.36Ile>Met) of the mature protein. In the LALBA gene, interesting polymorphisms at the promoter and exon levels were observed in South American Camels, potentially affecting gene expression and protein structure. The most notable polymorphisms were: g.112A>G, characterized by a positive impact on the LALBA promoter activity and its expression in alpaca milk and; g.1229A>G, resulting in an AA substitution (p.78Ile>Val) and two α -lactalbumin variants named A and B. Overall, this research provides significant insights into the genetic diversity and functional implications of milk protein genes in camels and offers a better understanding of camel genetics. Moreover, the identified polymorphisms offer potential applications for association studies with milk protein yield, facilitating future advancements in camel breeding and dairy industry practices.

Genome-wide Association Study of Hair Shape Variation in Dromedaries from Kuwait

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Within the frame of the Eleventh Illumina® Agricultural Greater Good Initiative, we designed and developed a medium-density Illumina® SNP array including 56K SNPs that is now commercially available. Genotypes from this subset of loci were obtained for a total of 22 Kuwaitis dromedary samples for which hair crimp type phenotypes were available (8 wavy vs 14 straight). Here, we present the preliminary results from a PLINK-performed case/control association test on the above-mentioned dataset, in order to identify genetic factors underlying the hair shape variation. Our analysis identified a supported genomic region on chromosome 3 spanning a range of about 157 kb and including three genes (HOMER1, TENT2 and CMYA5) putatively associated with the investigated phenotype. Of particular interest, HOMER1 emerges as a primary candidate due to its known involvement in the stimulation of the mTor/Akt pathway via a calcium-dependent mechanism. Notably, this pathway was found to be implicated in curly hair traits in other species, including the Mangalitzta pig, Chinese goats, and mice. These preliminary results provide novel insight into the molecular mechanisms underlying the hair shape variation in dromedaries. Further investigations are needed to validate the role of our candidate genes in determining the observed phenotype.

Session 26

Theatre 5

Case/Control Association Study on Trypanosome Infection in Algerian Dromedaries: Unraveling Genetic Determinants

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Among livestock species, dromedaries are particularly susceptible to trypanosome infections, resulting in substantial economic losses and compromising health and welfare of the affected animals, as well as a potential threat for human health. To date, the genetic factors underlying susceptibility or resistance to trypanosome infection in these animals remain poorly understood. We performed a preliminary case/control association study to identify genetic loci associated with trypanosome infection in Algerian dromedaries by leveraging genotyping and phenotypic data from 107 animals. The infection status had been defined based on PCR assays. The dataset was first pruned to extract only autosomal markers, then quality-checked by PLINK to filter out SNPs and individuals based on individual and locus missingness and minor allele frequency. Subsequently, we detected and removed a total of 13 outlier samples, thus obtaining a final working dataset including 80 animals (20 cases vs 60 controls) and 45,138 SNPs. Overall, we performed seven case/control association tests using different sub-samples of the control cohort. We identified several significant genomic regions including genes putatively associated with trypanosome infection. Out of them, two included the genes DIS3L2, COPS7B (chr5) and UBE2K (chr2) which were previously found to be involved in trypanotolerance in goats. These preliminary results provide novel insight into the genetic basis of trypanosome infection in dromedaries, paving the way for further studies to validate the observed signals.

Glycosylation dependent cell adhesion molecule, first genetic diversity investigation in dromedary camels
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Given its relatively lower selection pressure compared to other livestock species, the dromedary camel presents potential opportunities for further improvement in its economic and resilience traits. In this context, there is increasing interest in genetic investigation of economically related traits, particularly with the identification of genetic markers for future association studies. Therefore, the main objective of this study was to explore the genetic variation of the camel GlyCam-1 gene, which codes for the GlyCAM1, a major whey protein in camels. DNA from 10 dromedary camels was extracted from collected blood samples using the standard phenol-chloroform method. The entire gene and its promoter were first amplified and then sequenced using the Sanger method; followed by a bioinformatic analysis. GlyCam-1 gene consists of 4 exons and 3 introns, spanning 2379 bp. The Open Reading Frame (ORF) codes for 155 amino acids, and the translation stop codon is located between nucleotides 130-137 of exon 4. The polyadenylation signal (AATAAA) is also located in exon 4, 95 bp downstream from the stop codon. A total of 15 polymorphic sites were identified: 2 in the promoter region, 4 in the exonic regions and 9 in the introns. Among the identified single nucleotide polymorphisms (SNPs), the 4 mutations detected in the exons were synonymous (g.708C>T, g.744T>C, g.1485C>T and g.3008A>G). A preliminary analysis of transcription factors binding sites evidenced the influence of the SNPs g.234C>A and g.563 C>T on the transcription factors (TF). Further analysis is required to investigate the potential effects of all identified mutations on splicing process and expression levels.

Fragility and challenges in the development of the camel milk sector worldwide

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In less than 30 years, camel milk has evolved from a “gift economy” to a “market economy” in most camel countries. This important change contributed to the development of the camel milk production worldwide which has been multiplied by 9 since 1961 vs 2.3 for cow. However, with an estimated production of 4.12 million tons in 2022 (under-estimated), camel milk is representing 0.44% of the milk consumed worldwide (0.18% in 1961). Nowadays, on average 21.3 % of the camels are lactating (cows :18%). Moreover, the main part of the camel milk is still self-consumed by pastoralists: for example, 75% in Africa, 60% in Saudi Arabia. The introduction of camel milk into market is partly linked to climatic changes, to the necessity to provide milk in large cities from Middle East or Africa, and to the “health effect” attributed to this product. The consequences for the camel milk commodity channel were the trend to the intensification and “periurbanization” of the dairy camel farming systems, the development of milk processing for making long shelf-life products (as fermented, pasteurized or cheese) and the conquest of international market with the emergence of camel milk powder value chain. The development of a camel milk sector in western countries (USA, Australia, and Western Europe) has also to be emphasized. However, this development is fragile due to different factors: (i) the price differential with cow milk; (ii) the low milk productivity of camel; (iii) the weak structure of the commodity channels; (iv) the distance of production zones from consumption basin in many countries. Despite this fragility, a growth of the camel milk sector by 6-8% annually is expected for the next decennia.

Alpacas in Austria: Survey on current situation on management and breeding

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In Austria, there has been an increasing interest in alpacas in recent years. According to official data, around 10,000 animals (alpacas and llamas) are currently kept, and a further upward trend can be observed. Nevertheless, very little is known about the interests of the keepers, breeding objectives and general husbandry conditions. Therefore, this study aims to provide an initial overview of the current situation of alpaca production in Austria. Hence, an online survey was developed, and members of the Austrian Alpaca Association were invited to participate. The topics addressed were reasons for keeping alpacas, herd sizes, management practices, breeding strategies and questions related to the association. Out of 245 members, 101 persons completed the entire survey. 93 respondents indicated that they keep their animals as a secondary activity. Income is generated through sales of fibre products and breeding animals, but also activities (walking tours, etc.) were frequently mentioned. The majority keep Huacaya alpaca, and only ten respondents also have Suri alpacas. The herd size varies between 2 and 85 animals, with an average of 17. Body conformation, fibre quality and pedigree information are the three most important criteria for choosing a breeding male. 69% of the respondents confirmed their interest in participating in a structured breeding program and collecting systematic performance data, but only one-third were willing to pay for this service. The farmers keep up to date with the latest alpaca management news via the association and through seminars or online research.

Session 26

Theatre 9

Lessons learned from the intensification of milk production in dromedaries and how it advances the camel dairy industry in arid, semi-arid countries

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Camels are the fifth most important dairy animals and are an important food source in arid and semi-arid regions. Despite this fact, the intensification of the camel dairy industry only started 15 to 20 years ago. Such production systems are not widespread, although, it is expected to grow in the coming decades. One of the most important constraints is the lack of funding. Intensive camel milk production requires an initial investment, which has to include a continuous water and electricity supply, a regular and reliable feed source, as well as well-trained professionals and veterinary service, a processing facility and reliable access to markets. Unfortunately, today these requirements are difficult to meet in most arid and semi-arid countries where camels are kept naturally. However, if the above conditions are available, dromedaries can be integrated efficiently into an intensive production environment. Such a production can offer many advantages. Primarily, it allows the efficient and cost-effective production of high quality, raw camel milk that is suitable for further processing and meets the quality requirements of the consumers of the 21st century. At the same time, it also ensures that the animal health and animal welfare requirements of the species are met by complying to national and international guidelines, statutory requirements and standards. In addition, intensive production allows the development of selective breeding programs, the use of assisted reproduction and the establishment of so-called nucleus farms where high genetic potential dromedaries are concentrated. The experience gained in such farms can improve production in semi-intensive systems and can also enhance the breeding programs by using high genetic value males for mating. The aim of this presentation is to review our experience on the present status and challenges related to the intensification of camel farming in arid and semi-arid countries.

Study on anatomical structures of the dromedary udder

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With climate change and the scarcity of the food supply, dromedaries are becoming interesting food suppliers in non-arid regions of the world. Especially their milk, which is also described as the “white gold of the desert”, has excellent nutritional properties. The aim of the present study was to examine the mammary gland of dromedary camels using measurements, injection casts, frozen sections, ultrasonography, endoscopy and radiography. Ultrasonography and endoscopy are easy to perform in the field and feasible to diagnose pathological conditions of the mammary gland. For this purpose, 25 udders from camel carcasses, that arrived at CVRL in Dubai for pathological examination were examined. Also 49 udders were obtained from a local slaughterhouse for further examinations. These udders were analyzed using ultrasonography and radiography, with and without contrast medium, cast with injection of resin, gelatine or paraffin and frozen sections of native or casted udders were examined. Additionally, 11 lactating dromedary camels were selected for ultrasonographic udder examination. The injection casts and frozen section of the udder of the dromedaries shows a separation between the two teat canals and the two glandular complexes of each quarter. The structure of the teat and the parenchyma shows that the dromedary has specific anatomic structures which are important to know for application of machine milking and for improving udder health. The results of our examination showed that the ultrasonographic and endoscopic examination is a feasible tool for non-invasive diagnostics as well suitable for diagnostics in non-sedated animals. The examinations of camels restrained in a chute were easy to perform. However, additional examinations are necessary to evaluate pathological ultrasonographic and endoscopic images of the udder and teat of camels.

Session 26

Theatre 11

Insights into Dromedary Camel Pregnancy: Practical Applications of Dromedary Placenta Anatomy in Abortion Prevention Strategies

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Dairy farming of dromedary camels comes with significant challenges, including high abortion rates causing substantial production losses. Investigating the cause of these abortions is essential to achieve sustainable milk yields. Factors such as animal husbandry, nutrition, and epidemiology play crucial roles in their prevention, but despite optimal conditions in some farms, high abortion rates persist, encouraging focused research on other factors. The placenta serves as a portal for maternal-foetal nutrient and gas exchanges. Understanding this organ's composition and function is thus essential to improve pregnancy outcomes. Over the course of a year, we collected and studied aborted placentae and fetuses. We also included two uteruses of pregnant dromedary camel carcasses and studied the anatomy of these healthy placentae. This study provides a comprehensive description of the anatomy of the dromedary camel placenta, characterized by its diffuse, epitheliochorial nature, and goes over the main pathologies found as well as their impact on foetal development. Understanding the structures of the placenta is essential to grasp the adaptative mechanisms that enable camels to thrive in challenging habitats. Furthermore, this study compares the clinical picture of healthy placentae taken from dromedary carcasses with placentae obtained from aborted fetuses. Doing so, we aim to improve the understanding, diagnosis and prevention of abortion. This approach bridges the gap between anatomical knowledge and clinical practice, facilitating a deeper understanding of camel reproductive health management in arid regions. All samples were collected with the kind help and support of Central Veterinary Research Laboratory, Dubai, U.A.E and Camelicious, Dubai, U.A.E

Effect of pasteurization and simulated digestion on insulin content of camel milk

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Several factors may affect insulin (INS) content in camel milk. The objective was to evaluate the effect of pasteurization and simulated human digestion on INS content of camel milk. Milk pools representing early lactation (4 to 60 DIM), late lactation (180 to 210 DIM), and bulk commercial milk were used. Milk was pasteurized at 72°C for 15 sec and then placed on ice. Raw and pasteurized pools were exposed to stomachal and intestinal digestion (INFOGEST). INS was determined using an ELISA kit at 450 nm. The statistical model contained the effects of the pasteurization, milk pool, and digestion; the appropriated interactions; and the residual error. INS levels in early (0.57 µg/L), late (1.21 µg/L), and bulk (1.93 µg/L) milks differed ($P < 0.001$). INS values in raw (1.28 µg/L) and pasteurized (1.19 µg/L) milks also differed ($P < 0.05$). On average, INS values decreased ($P < 0.001$) from 1.24 to 0.23 µg/L during the stomach digestion. Although initial INS values were 8% lower in the pasteurized milk than in the raw milk, the concentration of INS remaining after the stomach digestion tended to be greater ($P < 0.10$) in pasteurized milk (0.32 µg/L) than in raw milk (0.14 µg/L). On average, only 19% of milk INS remained available for intestinal digestion. By the end of the intestinal digestion, remaining INS concentrations were neglectable (0.0003 µg/L; $P < 0.001$). Overall, pasteurization decreased INS concentration, but it protected INS during stomach digestion. Since INS disappeared during intestinal digestion, the antidiabetic effect of camel milk could be related to other bioactive peptides. Funded by the Saudi National Science, Technology, and Innovation Plan (Project #13-AGR1159-02).

Session 26

Poster 13

Adaptive evolution patterns in the genome of dromedary camels

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The genome of dromedary camels is a target of different evolutionary forces that may shape its diversity pattern. These forces are mainly attributed to the historical utilization of this species in transportation and trading, as well as their adaptation to the harsh environmental conditions they experience. In this research project, we have analyzed whole genome sequence data of dromedary camel populations from the Arabian Peninsula for two main purposes: 1) assess their genetic diversity and relationship, 2) investigate their genome for signatures of positive selection. To achieve the first goal, Principal Component and admixture analysis were conducted and revealed high genetic homogeneity among the dromedary populations analyzed. A degree of genetic distinction was also observed associated with their geographical distribution, with a substantial level of genetic admixture. Using the de-correlated composite multiple signals (DCMS) approach, which combines the signals from different analyses, a total of 36 candidate regions harboring 87 genes were specified to be under positive selection. A haplotype block and frequency analyses on these regions revealed a total of 185 blocks encompassing 1,340 haplotypes overlapping these regions. Approximately 2% of these haplotypes were found to be prevalent. These candidate regions, and their haplotypes, can be considered as primary favorable genetic elements in the dromedary genome that need to be conserved to maintain the genetic diversity of this well-adapted species.

Genetic parameters of fibre density in alpacas

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Fleece weight determines the income of the producer. Fibre diameter is the most commonly used selection criterion in alpacas, and this leads to an undesirable response in fleece weight due to the positive genetic correlation between them. However, higher fibre density increases the number of fibres compensating their lower weight, leading to heavier fleeces. Fibre density can be addressed by a subjective scoring method from 1 to 5, but also by a density quantification device (Fiber-DEN), where a 1 cm² area is shaved at the midline level and the skin dyed, differentiating very clearly the fibre follicles in white alpacas. This study aims to estimate the genetic parameters of hair duct number (HD), total fibre number (TF) and HD/TF ratio (RT) measured by Fiber-DEN and their relationship with the main textile traits such as fibre diameter (FD), medullation percentage (PM), greasy fleece weight (GFW), subjective density score (DE) and staple length (SL). A total of 402 fibre-DEN records of white Huacaya alpacas aged 1 to 16 years were used along with 8763 (FD and PM), 5752 (DE) and 11271 (GFW and SL) records for other traits. The pedigree consisted of 15 360 animals. The heritabilities were 0.40, 0.47 and 0.37 respectively for HD, TF and RT, and 0.31, 0.28, 0.16, 0.34 and 0.05 for FD, PM, GFW, DE and SL. The relevant genetic correlations between fibre-DEN traits and others were -0.50 for HD-FD and TF-FD, 0.36 and 0.43 respectively for GFW-TF and GFW-RT, and 0.50 for DE-GFW. DE and TF might be used as selection criteria to simultaneously reduce FD and increase GFW in a genetic improvement program, the latter being more costly

Session 26

Poster 15

Time budgets in stabled dairy dromedary camels

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Time budgets is used to determine the amount of time camels allocate to different behaviors throughout the day and can be a useful tool to evaluate animals' welfare and management systems. This work investigated the time budgets activities and their implications on the welfare of intensively managed dairy camels. Nine clinically healthy Maghrebi camels (aged 11.7 ± 1.9 years, weighing 437.8 ± 12.0 kg and 72.6 ± 7.1 DIM) housed in a loose stall barn were monitored for 6 consecutive days. Major behavioral activities such as standing, lying, walking, feeding, rumination, milking, as well as other activities like coprophagy, suckling, and sleeping, were continuously recorded. The mean duration of these activities was calculated to obtain the time budget. Results showed that camels spent most of their time in a standing position feeding ($P < 0.0001$). Specifically, camels were standing for 54% of their time and lying down for 38%. Data revealed that the main behavioral activities expressed were feeding (35.5%) followed by rumination (24.5%). Walking activity remained low throughout the day, representing only 2% of the total time budget of stabled camels. No common stereotypic behaviors were recorded; however, coprophagy was detected and recorded 10.3 ± 1.7 min/day. These results suggest that time budgets should be taken into consideration to improve management conditions and welfare status in stabled dairy camels.

Dairy cows' diversity of responses to heat stress: between resilience and adaptation

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In a context of global warming, there is a need for implementing multi-criteria evaluation of dairy cows' physiological and behavioral adaptation to chronic and acute heat stress to 1/ provide references and facilitate decision-making process in herd management and 2/ specify the phenotyping strategy in genetic selection. To this end, thirty-six Holstein cows in late lactation/gestation, producing 22.5 (\pm 1.5) kg/d of milk, have been monitored during summer in Western France. Environmental conditions were described with Temperature Humidity Index (THI). Behavioral and physiological indicators recorded were ruminal temperature, time spent panting, resting, ruminating, and eating, milk production and composition, plasma non-esterified fatty acids, cortisol, and thyroxine, and colostrum immunoglobulins. K-means clustering algorithms identified 3 groups of cows according to their response to acute heat stress (average daily THI superior to 74 during 4 consecutive days). Group A was composed of cows with low milk losses (-0,5kg/d) and a slight increase of ruminal temperature (+0,24°C), group B was composed with cows with low milk losses (-0,5kg/d) and a higher increase of ruminal temperature (+0,36°C) and group C was composed of cows with higher milk losses (-2kg/d) and higher increase of ruminal temperature (+0,36°C) and was characterized by higher milk fat and protein content during lactation. Group A, B, and C presented variable panting and/or eating/ruminating behavior adaptations. No difference in any of the other indicators measured was shown between groups. This project has received funding from the European Union's Horizon 2020 Programme for Research & Innovation under grant agreement n°101000226.

Session 27

Theatre 2

Genetic mechanism of brain-gonad axis regulating sex differentiation in sex-biased zebrafish families

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The hypothalamic-pituitary-gonadal axis is a well-known neuronal reproductive axis in vertebrates that triggers reproduction by activating the endocrine machinery. The brain is a key factor that combines environmental stimuli with physiological responses to direct gonadal fate in fish species whose sex determination is influenced by the environment. Changes in the expression of sex steroid hormones in response to environmental stressors lead to sex reversal, resulting in a male-biased (MB) or female-biased (FB) population. The zebrafish brain exhibits sex-specific differences at hormonal and transcriptional levels and is highly plastic even after sexual maturity. In this study, we investigated the transcriptional profiles of the zebrafish brain in an MB and an FB family exposed to high ambient temperature to gain new insights into the genetic mechanism of the brain-gonad regulatory axis that leads to sex ratio disequilibrium in response to elevated temperature. For this purpose, RNA-Seq was performed on brain tissue from 45 males and females in the control and high-temperature groups from two sex-biased families. The results showed a high number of significantly differentially expressed genes (SDEGs) in each gonad type in the MB versus FB family in the control group. Interestingly, the number of SDEGs was considerably reduced when the animals were exposed to high ambient temperature. A similar trend was observed in testes versus ovaries within each family type, with the number of SDEGs being higher in the MB family than the FB family in both temperature groups. Our study provides new insights into the importance of the brain-gonad axis as a transducer of environmental stimuli that cause sex ratio imbalance under rapid global warming.

Physiological and Productive responses to a natural short-term heat wave in Holstein Friesian and Brown Swiss lactating cows

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The aim of the study was to assess the different adaptative physiological and productive responses of Brown Swiss (BS) and Holstein Friesian (HF) lactating cows to a natural summer 4-days heat wave. Forty multiparous BS (n=20) and HF (n=20) dairy cows were involved, balanced for DIM, ECM Yield and BCS. The barn was provided with sprinklers and fans that were switched off for 4 consecutive days. The moment before turning off the cooling systems, at 4 AM was considered the thermal comfort condition (TC; THI = 63.4). The end of the 4th day, at 3 PM, was considered the heat stress condition (HS; THI= 81.3). Respiration rate (RR), rectal (RT), skin, eye, muzzle and vaginal temperatures were collected at 4 AM, 3 PM and 8 PM each day. Blood samples were collected at TC and HS. Biochemical, oxidative and inflammatory profiles were detected. Milk yield (MY) and quality were recorded. A GL Model was applied, considering as fixed effects breed and heat stress. RT increased each day from 4 AM to 3 PM in both breeds, and HF showed greater values ($P < 0.01$) during the whole trial. While these changes were more marked in HF, it is noteworthy that they are capable of more effective recovery and return to physiological levels during nighttime hours. BS did not change MY, whilst in HF cows MY decreased ($P < 0.05$) day by day. However, metabolic and inflammatory patterns did not show differences among the breeds. These results emphasize the necessity for additional research to comprehensively grasp the underlying differential mechanisms between the breeds.

Session 27

Theatre 4

Effects of Heat Stress on energy and protein metabolism in Iberian and crossbred pigs

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When pigs are exposed to heat stress (HS), their feed and water intake experience opposite adjustments, and their productive performance and protein and energy metabolism are reduced in order to maintain body temperature in normal ranges. In addition, the intense genetic selection focused on increasing lean tissue in modern breeds, aggravate this condition because of heat production (HP) associated with protein deposition, compared to other rustic and well-adapted breeds such as the Iberian (IB). Therefore, in order to analyze the mechanisms that allow the IB pigs to better adapt to HS conditions, we performed a 2×2 factorial design-experiment using 11 commercial crossbreeds (CO; Pietrain \times Landrace \times Large White) and 11 IB pigs, exposed to thermoneutral (TN; 23.5°C) and heat stress (HS; 33.5°C) temperatures during a total of 28 days. The last 6 days, pigs were housed individually in respiration chambers to accurately measure their feeding and water behavior, and nitrogen and energy balance from gas exchanges. Data were analyzed using the PROC MIXED procedure of SAS with temperature (T), breed (B) and their interaction ($T \times B$) as fixed effects. The pig was considered as experimental unit. As expected, preliminary results of this study show the adverse effects of HS on BW and growth in both breeds ($P < 0.05$), explained by lower feed intake (-1.23kg/d on average for the 2 breeds $P < 0.01$). This strategy to reduce HP (-360kJ/kg BW^{0.60} per day; $P < 0.01$) resulted in lower lipogenesis, as indicated from the decreased respiratory quotient under HS ($P < 0.01$). However, IB pigs had higher respiratory quotient indicative for a higher lipid synthesis in both TN and HS conditions than CO pigs. In addition, the increased water consumption during HS was less pronounced in IB (+5.7L/d) than in CO pigs (+15.5L/d; $P = 0.01$), which also resulted in much lower urine production. This research was supported by the Spanish Ministry of Science and Innovation (grant PID2021-127949OB-100). The PIGWEB project has received funding from the EU Horizon 2020 research and innovation program (grant agreement No 101004770).

Can we breed cattle with more efficient rumen microbiome?

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Milk and meat from ruminants have reduced malnutrition and improved prosperity in many societies. However, ruminant production is facing significant challenges in terms of improving feed efficiency (FE) and reducing its environmental footprint. Recent research has revealed that variations in the composition and function of the rumen microbiome can directly and/or indirectly influence several phenotypic traits in cattle such as FE, methane emissions, metabolic health, milk and meat quality. This presents an opportunity for rumen microbiome interventions to improve productivity, health and reduce the environmental footprint of cattle production. More and more evidence revealed the individualized rumen microbiome and host genetics could be one of the factors influencing the colonizing of selective rumen microbes. Here, we reported the identified heritable rumen bacteria in both beef and dairy cattle. Some of the traits of the host that influence the composition and function of the rumen microbiome appear to be heritable. The extent of this heritability is also influenced by host genetics and has implications for cattle production-related traits. Moreover, we showed that heritable rumen bacteria exhibited a high degree of maternal similarity as compared to those that were nonheritable. Our findings illustrated that both the host and rumen microbiome cattle production and such traits associated with the rumen microbiome could be passed from parents to offspring. The identified genotypes may potentially serve as markers for breeding cattle for optimal rumen function.

Session 27

Theatre 6

Characterization of physiological responses in Rasa Aragonesa ewes under water restriction

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The aim of this study was to characterise the effect of water restriction on ewes. Two hundred and two Rasa Aragonesa ewes were subjected to total water restriction for 5 days. Intake, body weight (BW) and body condition score (BCS) were measured daily during the 5 days of restriction, as well as temperature and percent humidity. Blood samples were collected just prior to the challenge (0d), and at the end of the study (5d) for haematological and metabolite measurements. Wool samples were taken at day 0 and 4 weeks later (28d). Variation in blood and wool traits was also calculated for each animal. Dry matter intake, BW and BCS were decreased during the trial. Mixed model statistical analysis indicated that all haematological and metabolite levels changed during the water restriction period except for plasma cortisol. Principal component analysis (PCA) and hierarchical clustering were performed with variation traits between 0d and 5d (blood samples) and 0d and 28d (wool samples) to identify different responses among ewes. Four clusters were identified: 1) fat mobilization ewes (n= 56; wool cortisol increase and low increase neutrophils-to-lymphocytes ratio (NLR)) with low dehydration but highest fat mobilization; 2) high-stressed ewes (n= 9; very high plasma cortisol increase) with low dehydration; 3) low-stressed ewes (n= 106; low plasma cortisol and low NLR increase) with minor fat mobilisation but high dehydration; 4) stressed ewes (n= 31; high NLR increase) with higher dehydration. In general, the Rasa aragonesa ewes exhibit a high tolerance to water stress (C1 & C3 = 162 ewes; 80%) but with different adaptation responses.

Links between gills genes expressions during moderate heat stress and resistance to acute hyperthermia using rainbow trout isogenic lines

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In aquaculture, acute hyperthermia resistance is a trait of interest in climate change context. However, acute hyperthermia resistance, measured in laboratory conditions as time to loss of equilibrium when exposed to high temperature, is a trait whose underlying physiological mechanisms are not fully understood. It is important to understand these mechanisms to assess the usefulness of acute hyperthermia resistance trait as a predictor of fish ability to adapt to climate change. In the present study, we investigated links between resistance to acute hyperthermia and gene expressions in gills of rainbow trout exposed to heat stress. For this purpose, we phenotyped 6 rainbow trout isogenic lines for acute hyperthermia resistance and 82 genes gills expressions. Expressions were analysed before and after exposition to moderate heat stress (11°C to 23°C in 1H30 then 30 min at 23°C). Genes studied were chosen to represent several physiological functions such as metabolism, immunity, etc. Genes whose expressions were significantly differently expressed between isogenic lines resistant to acute hyperthermia and isogenic lines sensitive to acute hyperthermia were considered as genes potentially involved in acute hyperthermia resistance. Resistant lines differential gene expressions between the beginning and the end of the heat stress were higher for genes associated with O₂ transportation and innate immunity and lower for those associated with active ion transportation and gill permeability. This suggests that energy allocation during heat stress is different between resistant and sensitive fish.

Time-dependent molecular adaptation of immune and platelet function to heat stress in PBMC of Holstein dairy cows

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Dairy cows adapt to environmental heat by reducing feed intake, milk production and increasing peripheral blood flow to the skin. However, there is evidence that chronic heat initially stimulates a systemic inflammatory response that impairs the immune competence of dairy cows. The study aimed to elucidate the time-dependent effects of moderate chronic heat stress in peripheral blood mononuclear cells (PBMC) of Holstein cows. Twenty German Holstein cows (166±11 days in milk) were kept in a climate chamber under thermoneutral conditions (16°C; 63% relative humidity (RH) with a temperature-humidity index (THI) of 60 for 6 days (d) and fed a total mixed ration ad libitum. Subsequently, 10 cows were exposed to 28°C (heat stress – HS; with 52% RH; THI 76) for 6 d, whilst 10 cows were further kept at thermoneutral conditions (CON). Blood samples were collected at 24 hours (h) and at 6 d after HS for PBMC isolation. Isolated RNA was utilized for RNA sequencing (Illumina HiSeq2500) and analyzed for differentially expressed genes and pathway enrichment analysis (KEGG). Comparing 24 h versus 6 d revealed an upregulation of pathways related to platelet activation, fluid shared stress and arteriosclerosis which may be related to HS-induced microvascular injuries or changes in hematocrit as a consequence of prolonged HS. The activation of leukocyte transendothelial migration, ECM-receptor interaction, neutrophil extracellular trap formation and HIF-signaling pathway may indicate an activation of the peripheral immune response and oxygen delivery to maintain homeostasis after prolonged heat stress.

How does ewe age affect reproductive performance relative to rainfall conditions?

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Livestock producers will have to explore various management and mitigation strategies to maintain production levels in the face of adverse weather conditions and a changing climate. It is well-documented that ewe age affects reproductive performance, but it is unclear whether this phenomenon can be utilised to mitigate adverse environmental effects. This study was therefore carried out to determine whether certain age groups are better adapted to reproduce under specific rainfall regimes. Historical rainfall data collected on Tygerhoek Research Farm, Western Cape, South Africa between 1975 and 2018 was used to classify years as dry, below average, average, above average and wet. Ewe performance for conception, number of lambs born, number of lambs weaned, average birth weight and average weaning weight was calculated for each age group (2-6 years) across all years and within rainfall classes. Age group performance for each trait in each rainfall class was then compared to the overall trend. Overall, reproduction rate was maximal at three to four years of age and this trend was largely consistent across rainfall classes. Weaning weight was also highest in three- and four-year-old ewes. Birth weight increased with age, except in wet years, when no discernible trend was present. In dry years however, there was a clear trend for weaning weight to increase as ewes aged. Hence it is concluded that flock age structure cannot be used as a tool to mitigate environmental impacts if the age structure is already optimal for reproduction.

Session 27

Theatre 10

Sperm mitochondrial gene-dependent activity mediated the stress capacity in young and old Holstein bulls

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Mitochondria are key for stress resilience as all types of stresses are found to influence the mitochondrial DNA copy number (mtDNA-CN). However, the cross-talk between mtDNA-CN and stress tolerance in spermatozoa remains unclear. Therefore, this study aims to identify the mtDNA-CN and the expression levels of all mtDNA protein-coding genes (13 genes) in bovine sperm cells throughout the year. Accordingly, the semen from four old and four young Holstein bulls was collected during the four seasons. The DNA and total RNA were isolated from sperm cells. Mitochondrial DNA copy number (mtDNA-CN) was performed using a qPCR-based method using the isolated DNA. The RNA was subjected to cDNA synthesis followed by the mRNA expression analysis using qPCR. The data were statistically analyzed using the Student's t-test and one-way ANOVA analyses. Our findings revealed that compared to old bulls, the mtDNA-CN of young bulls was relatively higher in winter and summer but lower in spring and autumn. At the transcriptional level, old bulls' spermatozoa exhibited relatively higher expression levels of the ND4L, ND6, COX1, and CYTB genes in winter and the ND4 gene during the spring. However, young bulls' spermatozoa showed high expression levels for the ND1, COX2, ATP6, ND3, COX3, and ND2 genes in winter, and for the ND1, ATP6, ND4L, ND5, COX3, and ND6 genes in spring. Moreover, the ND1, COX2, ATP6, ND4L, ND4, ATP8, ND3, COX3, and ND6 genes in summer, and the ND1, ATP6, ND4L, ND4, COX3, ND6, COX1, ND2, and CYTB genes in autumn were upregulated in young bulls' spermatozoa. In conclusion, sperm mtDNA-CN indicated a correspondence with differential expression patterns in a gene-dependent manner in old and young bulls' sperm cells.

Modeling the changes in daily rumination time and milk yield to detect the onset of heat stress in lactating dairy cows

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Determining the temperature-humidity index (THI) threshold for the onset of heat stress (HS) can serve as a decision-making tool in HS management in dairy cows. While HS thresholds are usually based on milk yield (MY) losses, other physiological changes may appear at lower THI. The study aimed to determine HS thresholds of daily rumination time (RT) and MY of cows based on THI according to parity and lactation stage. Daily RT and MY were collected from 915 Chinese Holstein cows [533±74.7 min RT/d; 37.9±8.93 kg MY/d (mean±SD)]. Daily THI in the barn ranged from 25.6 to 84.8. Two data sets were used to compare parity (PRIMI or MULTI), and, only for multiparous cows, lactation stage (EARLY, PEAK, MID, or LATE at 1-40, 41-60, 61-160, or 161-305 days in milk, respectively). The effect of daily THI on RT or MY was analyzed using a generalized additive mixed model. Pointwise slopes of the fitted means were calculated. The threshold was considered as the THI when RT or MY started to decrease ($P < 0.05$). Above THI 58.8, RT decreased for PRIMI and MULTI by 3.56 and 4.35 min/d/unit THI, respectively). Only MULTI decreased MY above THI 65.0 (-0.078 kg/d/unit THI). As for lactation stage, EARLY, PEAK, MID, and LATE decreased RT above THI 57.0, 57.6, 59.4, and 61.7 by 3.89, 4.08, 5.02, and 3.14 min/d/unit THI, respectively. Only MID decreased MY above THI 66.4 (-0.195 kg/d/unit THI). Management strategies should be applied before THI exceeds 57-59 since HS can already have negative effects on the cows even without apparent production losses.

Session 27

Poster 12

Rumen-protected dry grape extract supply during natural heat stress improves the whole-blood immune response of Simmental cows

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Heat stress causes detrimental effects on the immune system and affects inflammation of dairy cows. To mitigate this negative response, recently different nutritional strategies were adopted. We aimed to assess the effects in mid-lactating Simmental cows of NorGrape® BPO (a rumen protected dry grape extract rich in water-soluble polyphenols) on immune response during heat stress. Thirty cows were blocked by DIM, parity, and MY and randomly enrolled in 2 groups: treated (BPO; 15) and control group (CTR; 15). In the BPO group, each cow received 470 mg/d of NorGrape mixed with corn meal and top-dressed onto TMR for 35 d during summer. Blood samples were collected at 0, 17, 31 and 35 d for the inflammatory and oxidative stress response and at 35 d for phagocytosis. Data were analyzed by PROC GLIMMIX of SAS. Circulating neutrophils and monocytes were higher in BPO cows than CTR cows after 35 d of treatment ($P < 0.05$). In addition, neutrophil phagocytosis efficiency was greater in BPO than CTR cows at 35 d (39.32 vs 32.08±2.32%; $P = 0.03$). Likely due to the greater phagocytosis, BPO group had levels of plasma advanced oxidation protein products than CTR group ($P < 0.05$). These results support the use of grape extract rich in water-soluble polyphenols as effective approach to help mitigate oxidative stress in heat-stressed cows and to enhance a better immune response through an improved neutrophil function and higher circulating neutrophils and monocytes.

Individual variability in trajectories of key plasma biomarkers involved in body reserves dynamics in meat ewes

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The mobilization and accretion of body reserves (BR) constitute one of the key mechanisms in ruminants for resilience and adaptation. A better understanding of inter-individual variability for candidate proxies of such mechanism is required before their use in breeding strategies. Primiparous and multiparous ewes were monitored during successive productive cycles in two contrasting farming systems (FS, indoor vs extensive) at five key physiological stages (Mating; mid-Pregnancy; 2 weeks Pre-Lambing; 3 weeks Post-Lambing; Weaning) for plasma biomarkers (NEFA, non-esterified fatty acids; BHB, β -hydroxybutyrate; T3, Triiodothyronine; INS, insulin). Unsupervised cluster analysis was done to investigate the variability in BR biomarkers trajectories. Overall mean trajectories for biomarkers followed BR dynamics throughout cycles and were mainly characterized by increases and decreases in NEFA, BHB, T3 and INS levels during BR mobilization and accretion phases, respectively. For each biomarker, two to three clusters of ewes were found depending on the parity and/or the FS. Trajectories between clusters differed in the levels and/or the shape. One of the major discrepancies between clusters was levels and/or time point when peaks of biomarkers were observed during BR mobilization. Higher individual variability in biomarkers was thus particularly observed around lambing. Genetic contribution in such variability for BR biomarkers was also investigated before considering these traits for breeding purpose. [Funding with H2020 iSAGE project 679302].

Differences Between Swiss Dairy Breeds in the Relationship Between Milk Urea Nitrogen and Urine Nitrogen Excretion

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The aim of this study was to model the major substrate for ammonia emissions, urinary nitrogen excretion (UN; g/day), based on an easy-to-determine proxy. One such proxy could be the milk urea nitrogen content (MUN; mg/dL). We therefore investigated urinary nitrogen and milk urea nitrogen values from 21 different feeding experiments performed with 162 Brown Swiss (BS), 348 Holstein (HO), 44 Jersey (JE) and 50 Swiss Fleckvieh (SF) cows. The relationship between the coefficients of UN and MUN was investigated using linear mixed models. The final model included the fixed effects MUN, body weight (BW), breed and the interactions MUN \times breed and BW \times breed, as well as experiment as random effect. The model also considered the heteroscedasticity. The coefficient of determination of the final model was high ($R^2 = 0.812$). The regression coefficients and highest posterior density intervals for MUN were 7.28 (5.42;9.02), 10.41 (9.16;11.65), 9.00 (6.20;11.94) and 11.79 (9.15;14.25) g/day for the UN increase per mg/dL for BV, HO, JE and SF, respectively. The estimated regression coefficients for BW and highest posterior density intervals were 0.09 (-0.01;0.19), 0.17 (0.11;0.27), 0.51 (0.21;0.80) and 0.28 (0.03;0.53) for BV, HO, JE and SF, respectively. The estimated marginal means \pm standard errors for UN (g/d) were 117 ± 12 , 147 ± 12 , 119 ± 14 and 135 ± 14 for BV, HO, JE and SF, respectively. These results are consistent with other studies and show that the MUN content of the milk, to be determined in tank milk samples, is a useful tool for predicting UN in lactating dairy cows. However, the results also show that the predictive model must consider breed and BW. This procedure enables improvement of the accuracy of mass flow models to estimate ammonia emissions which are based on the nitrogen excretion of livestock at the beginning of the manure management chain.

European Network on Livestock Phenomics (EU-LI-PHE): boosting phenotyping in livestock

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Phenomics in livestock is focused on the systematic description of the animal phenome, referred to as the physical and molecular traits of an animal. EU-LI-PHE is a Europe-centred multidisciplinary, interconnected and inclusive network of experts aiming to boost scientific collaboration, catalyse developments, and transfer of livestock phenomics concepts and applications to improve the sustainability of the European livestock production sector. EU-LI-PHE is focused on i) phenotyping technologies and infrastructures for applications in livestock phenomics, ii) novel approaches and methods for genome to phenome integration in livestock species, iii) computational resources and data analysis methods needed for this big data discipline, iv) the regulatory framework and the societal vision for livestock phenomics and v) the development of a training environment for the benefit of the next generation of researchers in this field.

Session 28

Theatre 2

Structured resilience phenotyping using big dairy data

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Part of the challenge in breeding for increased resilience is defining the phenotypes of resilience traits in livestock. Resilience indicators based on lactation curves are being developed and their genetic basis is under investigation. Increased adoption of robot milking systems and other smart farming equipment means that a broad spectrum of real-time cattle data is produced on modern dairy farms, which can be combined with regional weather data to discover phenotypes of climate response in dairy cattle. The SLU infrastructure for cattle data, Gigacow is continuously gathering PLF data including feed intake, milk yield, and health events from 15 Swedish commercial dairy herds since 2019. This information is crosslinked per cow with the national cow database Kokontrollen, and 50k SNP genotypes from the Nordic Cattle Genetic Evaluation. Uppsala University students in collaboration with SLU used SLU Gigacow milk yields normalized against Wood's Lactation Curve in generalized additive models to show negative impacts on milk yield from increased mean temperature and mean THI. With SLU Gigacow lactation curves and corresponding weather data, it is possible to detect depressed lactation in individual cows, and link it to genotype by GWAS. This serves as a proof-of-concept of a systematic approach to livestock genotyping, similar to the process in human disease genetics. Automated data collection produces signals, which are associated with genetic factors, and a structured phenotype can be defined, including the data sources necessary to detect it and investigate its heredity. A structured phenotyping approach would both accelerate and empower resilience research to fully utilize the expanding range of data provided by modern smart farms.

Large-scale phenotyping through milk infrared spectroscopy for enhancing resilience in dairy cattle: the contribution of LEO 'Livestock Environment Opendata' project

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This study aimed to explore the potential of the Fourier-transform infrared (FTIR) spectra stored during milk recording schemes for large-scale phenotyping of blood biomarkers associated with metabolic stress and resilience, including hepatic enzymes, ceruloplasmin (CP), total reactive oxygen metabolites (ROMt), and paraoxonase (PON). Blood metabolites were predicted (p) using FTIR spectroscopy with equations previously developed on an independent calibration dataset (1,351 cows, 5 herds). Calibration equations were then applied to a population database of 1,355,730 test-day FTIR milk spectra (176,174 cows, 966 multibreed farms), collected by the Breeders Association of Emilia Romagna region (ARA, Reggio Emilia, Italy) and made available by the Italian Breeders Association (Rome, Italy) through the LEO project. All predictions were analyzed using a linear mixed model. Days in milk was a relevant source of variation for the traits of concern ($P < 0.01$), with pPON showing opposite direction over lactation compared to pCP and pROMt, thus confirming their contrasting effects on inflammation/oxidation status. Traditional mountain farms exhibited higher levels of pPON compared to 'modern' farms with total mixed rations ($P < 0.01$). About breed effect, Holstein Friesian cows displayed the highest levels of pPON and lowest level of pCP and pROMt across all farming systems. These new phenotypes are promising for animal welfare monitoring and breeding purposes to address metabolic stress and improve resilience in dairy cows.

Session 28

Theatre 4

Ensemble Learner Enhances Prediction Accuracies for Extreme Body Condition Classes in Jersey Dairy Cattle in a 3D Image High-Throughput System

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High-level management of dairy herds relies on monitoring the body condition, but routine evaluation is limited, prompting exploration into cost-effective alternatives. We aimed to establish a reliable body condition prediction for Jersey cows with input from the 3D camera system CFIT (Viking Genetics, Denmark). We collected 2,167 body condition score (BCS) records from 794 Jersey cows. In total 691 prediction features were derived from top-down 3D images, defining contours on the cow's back. We implemented two data splitting approaches: 1) 7:3 random split, 2) herd cross-validation. Using LazyRegressor in Python, we observed Bayesian Ridge and Lasso Regression were the two top model choices. The models were tested with and without weights on class frequencies, and an ensemble prediction was formed by combining weighted and unweighted predictions. Evaluation metrics were F1-Score and accuracy on exact and 0.5-unit deviation, using rounded predictions. A linear model ($BCS = \alpha + \beta \times \text{predicted BCS}$) was used to evaluate bias (α) and inflation (β). The ensemble learner outperformed the component models. In the 7:3 split, overall accuracy reached 55.1% and 96.5% for exact and 0.5-unit deviation. F1-Score was improved in the highest BCS classes (exact: 0 to 31%; 0.5-unit: 0 to 68%). The ensemble learner exhibited superior performance in α and β (0.09 & 0.97) compared to the component models (α : 0.55-0.87; β : 0.68-0.82). Herd cross-validation showed a modest ~6%-unit reduction in prediction accuracy. Our findings shows the feasibility of predicting BCS in Jersey cows using a 3D camera system, with the potential to enhance management decisions in Jersey cows.

Phenotyping of beef calves BW and morphological scores based on 3D imaging and AI

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Into the PHENO3D project, we validated a new 3D device adapted to high-throughput on-farm phenotyping (Do et al., 2024). This enabled development of an AI that predicts weight and linear scores from the 3D image provided by the scanner. The linear scoring resulted on 2 synthetic scores: the muscular conformation (MUS), and the size of the animal relating to skeletal development (SKE). Currently, these linear scores are done visually by certified technicians. To achieve these objectives, 1194 Charolais calves from 4 to 12 months and from 90 to 620 kg were scanned in 14 farms. Most of them were scanned 2 times resulting in 2110 3D images. Reference measures were collected on animals: each calf was weighed (BW on an electronic scale) and linear scored by 3 trained technicians. For BW and linear scores prediction, 1818 and 1575 images were respectively split into train (80%) and test (20%) subsets. For BW, MAE was 12.1 kg (4.2%). The quality of the linear scores' prediction was assessed in the same way that current technicians are yearly evaluated: a spearman correlation with a reference (homogeneity), a spearman correlation between two scoring sessions (reproducibility). For SKE, the homogeneity and reproducibility were respectively $r=0.76$ and $r=0.87$. For MUS, the homogeneity and reproducibility were respectively $r=0.79$ and $r=0.81$. The performances of the linear scoring models are superior to the average performance of the technicians in their yearly certifications. The performances of the AI on the Charolais breed gives the green light to multiply our models on the 9 other beef breeds linear scored today.

Molecular phenotyping through metabolomic analyses in pig breeds

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The animal phenome can be defined as the ensemble of the physical and molecular traits of an animal. The molecular traits can be further dissected into different layers of internal phenotypes, also including many metabolites. Here we analysed ~800 metabolites detected from an untargeted metabolomic platforms on plasma of ~700 Italian Large White and ~300 Italian Duroc pigs to identify metabolites that could be useful to characterise the two breeds. Animals were sib-tested pigs, included in the selection programmes of Italian heavy pigs run by the Italian Pig Breeders Association (ANAS), raised in the same genetic station, and slaughtered at nine months of age when they reached about 160 kg live weight. After quality control, univariate, multivariate and machine learning analyses were used to identify metabolites that could discriminate the two breeds. Combining information from these approaches, a total of ~100 metabolites were identified (including ~1/3 amino acids and ~1/3 lipids among others). The selected metabolites clearly separated the two pig breeds. The observed differences for these molecular phenotypes can describe genetic differences between Italian Large White and Italian Duroc pigs. Acknowledgments: This study has received funding from the European Union's Horizon Europe research and innovation programme under the grant agreement No. 01059609 (Re-Livestock project).

Integrating genomic information in metabolomic networks to dissect molecular phenotypes in pigs
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The complexity of an organism arises from the interplay of different molecular layers (e.g. the genome, transcriptome, proteome, and metabolome) that generate biological processes and pathways that, in turn, define the animal phenotype. In this study, we combined metabolomics and genomics to explore and gain knowledge on the interplay between these two layers using systems biology approaches. For these aims, we used both genome (~60,000 SNPs) and plasma metabolome information (~1,000 metabolites obtained from targeted and untargeted platforms) from ~1,000 Italian Large White and Italian Duroc pigs. Metabolomics data were used to reconstruct pathways through two network generation approaches, i.e. correlation networks and Gaussian graphical models. Then, genome-wide association studies were carried out to identify genetic loci influencing metabolites (mQTL). A first comprehensive catalog of mQTL was obtained, including a few hundreds of putative causative genes. These loci were subsequently studied and used to inform metabolite networks. The inclusion of mQTL in the metabolic pathways improved the estimation and generation of simple correlation networks and revealed relationships between known and unknown metabolic features. These results provided a first picture of genetic factors and metabolic interactions affecting the pig metabolism. Acknowledgements: This study has received funding from the European Union's Horizon Europe research and innovation programme under the grant agreement No. 01059609 (Re-Livestock project).

Novel resilience indicators based on egg production in laying hens

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Breeding for heat tolerance aims to minimize the impact of heat waves on production and welfare of livestock. Here, we aimed to define new phenotypes based on perturbations in egg production of laying hens, which can be later used as indicators for heat tolerance. We analysed 1.8 million daily egg production records of laying hens housed in 4,806 cages on two farms over approximately one year. We quantified perturbations at cage level as the deviation of daily egg laying rate (LR, %) from the expected laying rate, where the expected laying rate of each cage was estimated using the Adams-Bell curve. We estimated the phenotypic correlations of several potential resilience indicators, including the natural logarithm of the variance (LnVar) of the LR, cumulative sum of the downward deviations from expected production, and the persistence of egg production. Between 35 and 60 weeks of age, higher variance (expressed by LnVar) of the egg laying curve was phenotypically correlated with larger cumulative deviations from expected production ($r=0.717$), less eggs produced ($r=-0.445$), and lower persistence of egg production ($r=-0.397$). That is, more resilient laying hens (i.e., hens with more consistent egg production over time) produced closer to their expected production, laid more eggs, and maintained high production for a longer period of time. We also observed larger deviations from the expected laying rate at higher indoor temperatures, but weather and production stage effects were not clearly separable in the dataset at hand. In the next step, we are going to link these novel indicators to heat tolerance using the data of multiple farms over several years.

Towards a 24hr chicken surveillance system

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Increasing demands for animal welfare in livestock production require the inclusion of behavioral data into future laying chicken breeding programs. This is accompanied by challenges of data recording in cage-free layer systems and increasingly labor-intensive requests for animal control by authorities, which settles the raising need for automated 24hr animal monitoring systems. RFID based chicken tracking was shown to allow for accurate animal identification, but at low resolution and thus biasing measures of animal movement. In contrast, computer vision models allow accurate detections of moving animals, but tracks are usually lost at points of occlusion or disturbances, thus hardly allowing animal identification over a long period. To overcome those limitations, we currently develop a combined real-time monitoring system through video tracking and animal identification based on sparsely placed RFID antennas for our research flocks. For an initial test set-up, three low-frequency RFID antennas (30*80cm) were placed in a pen on two levels, stocked with eight hens and a rooster. Chickens were detected, using a custom-trained YOLOv8n (640*640px) object detection model based on 10fps videos (F1 score of 0.96 at a confidence of 0.8), followed by assigning tracks to the predictions by SORT algorithm. Further, the most likely chicken was assigned to the tracks by minimum average squared distance to antennas with RFID signals. This resulted in increasing the average length of tracks from 1.9min (47min max) to an average track length with an id assigned of 8.2min (153min max). However, using the Rooster as a positive control currently results in 48% correct classifications only, thus requiring further updates of the assignment algorithm in identification of track switches and closing of gaps between assigned tracks. Nevertheless, those intermediate results are promising to soon allow for an accurate 24h monitoring of laying chickens, which will leverage the information of future research trials.

Session 28

Theatre 10

A tracking tool for monitoring activity in broilers

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An important percentage of the resources used by a growing animal is spent on physical activity. It has been shown that selection for reducing residual feed intake has led to less active animals. Our purpose with this communication is to inform the poultry researchers community about the development of a new computer vision system to track and quantify the physical activity of chicken. We have used MaskR-CNN library to perform the instance segmentation in order to detect all the visible chickens in frame, MaskR-CNN produces a table with the coordinates of each chicken. In a second step, the detected chickens were tracked across frames using Simple Online and Real-Time Tracking (SORT). This initial demonstration was done with a mobile phone camera, recording 20 one-minute videos of 848 x 478 pixels at 30 frame per seconds (fps), at a high of approximately 1.70 meters. The animals were in a commercial broiler farm with a total of 17,000 Ross broilers with an approximated density of 17 animals per square metre. For the detection a model trained on images from rabbits was used as starting point, this model was re-trained just with 16 frames on which all the observed chickens were annotated, 50-60 individuals per image. Three additional images, having approximately 55 chickens each were reserved for validating the detections. The mean Average Precision of pixels assignation to the chicken class in the validation set was 0.92. Due to this high accuracy, tracking performance was high even when videos were cut to 10 fps, but the performance was considerably better when the quality of the videos was maintained at 30 fps. These results indicate that group tracking to measure physical activity of a group of chicken is feasible even at high density, further training rounds and a proper definition of physical activity metrics are needed

Computer-vision based analysis of turkey locomotion

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Accurate measurement of livestock locomotion is crucial across various domains, including optimizing herd management practices, early detection of health issues, and understanding behavioral patterns for enhanced productivity. However, conventional data collection methods encounter limitations in terms of cost and scalability. Recent advancements in computer vision offer a promising solution for large-scale, automated, non-intrusive, and cost-effective phenotyping of locomotion. In our study, we implemented a video-based system to track over 300 turkeys within a 120 m² pen, employing multiple cameras covering the entire area. We segmented days of videos into five-minute clips and randomly sampled 280 clips to derive locomotion parameters. First, we applied our previously developed multi-camera tracking algorithm to track the turkeys. Second, we derived locomotion parameters from the detected tracks. This included estimating the speed and turning angle distributions of individual turkeys, and assessing the time each turkey spent on resting (defined as speeds < 0.5 m/s lasting for more than 2 seconds) or moving. Additionally, we determined the time each turkey spent on feeding based on detecting its RFID-tag (Radio Frequency Identification) in the feeder. Furthermore, at the population level, we analyzed the distribution of animals within the pen, offering insights into social dynamics and space utilization. The results revealed significant individual differences in locomotion parameters and time budgets. Notably, despite being kept in the same environment, the results suggested that these traits may have a genetic basis. Overall, we developed a video-based approach for locomotion measurement and found substantial individual differences in locomotion parameters and time budgets among turkeys. Consequently, we gained valuable insights into individual behavior, social dynamics, and space utilization, facilitating informed and effective livestock management and breeding practices.

Session 28

Theatre 12

Inertial measurement unit technology for gait detection: a comprehensive evaluation of gait traits in two Italian horse breeds

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The shift from agricultural to leisure and sports purpose of the horse breeding sector led to a decrease in local breeds' population size, due to a loss of their original breeding purposes. Most of the Italian breeds thus must adapt to modern market demands, and gait traits are suitable phenotypes to help this process. Inertial measurement unit (IMU) technology can help to objectively detect them. This work aims to investigate gait traits based on an IMU sensor through i) the evaluation of which environmental factors and biometric measurements influence the IMU measures, ii) the repeatability, and iii) the correlation with judge evaluations. The Equisense Motion S® was used to collect phenotypes on 135 horses, Bardigiano (101) and Murgese (34) and the data analysis was conducted using R (v.4.1.2). Analysis of variance (ANOVA) was employed to assess the effects of biometric measurements, and environmental factors on the traits. Variations in several traits depending on the breed were identified, highlighting different abilities among Bardigiano and Murgese horses. Repeatability of horse performance was assessed on a subset of horses, with regularity and elevation at walk being the traits with the highest repeatability (0.63 and 0.72). The positive correlation between judge evaluations and sensor data indicates judges' ability to evaluate overall gait quality. The predictability of judge evaluations was assessed via a support vector machine model (SVM). High accuracy (ranging between 0.62 to 1.00) was shown, despite challenges posed by an unbalanced number of observations per subjective classes of evaluation and a limited number of animals. In conclusion, integrating IMU technology offers valuable insights into horse performance evaluation, with implications for breeding and training. Michela Ablondi is co-financed by the European Union – NOP Research and Innovation 2014-2020 art. 24, par. 3, A) Law 30/12/2010, n. 240 and of the D.M. 10/08/2021 n. 1062.

LIPH4SAS (« Livestock Phenotyping for Sustainable Agroecological Systems »): a national research infrastructure for livestock phenotyping

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The LIPH4SAS research infrastructure is dedicated to livestock phenotyping and enables experiments, phenotyping and biological sampling for the animal research community, with a view to promoting the transition to more sustainable agricultural and food systems based on agroecological principles. LIPH4SAS comprises eight experimental units, of which four are dedicated to ruminants, two to pigs and two to trout. It also includes a platform dedicated to the in-depth study of livestock physiology and a structure responsible in charge of data management and the development of phenotyping tools. LIPH4SAS has large facilities (2,000 cattle, 4,500 sheep/goats, 7,000 pigs ...), including many valuable genetic resources (original populations, selected lines, homozygous trout lines, etc.), managed in a variety of geographical areas and breeding systems. It provides skills and tools for detailed and multi-scale phenotyping (surgery, in and ex vivo imaging, respiratory chambers, etc.) and horizontal phenotyping on large groups of animals (slaughterhouses, equipment for measuring growth, feed intake and efficiency, body composition, behavior and health, product quality, greenhouse gas emissions, etc.). The aim of LIPH4SAS is to provide a global service that promotes interaction between scientific communities, harmonizes practices to ensure the highest level of expertise as possible and ethics in animal experimentation and welfare, encourages innovation in animal science and facilitates access to Fair data.

Session 28

Poster 14

Exploring the potential of an image analysis mobile app to estimate horse body morphometric measurements

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The ability to measure an animal directly from images without physically taking measurements could prove to be a valuable tool for studying its growth pattern and morphometry. This study aims to employ an image analysis mobile app to estimate body measurements (BM) from pictures. A total of 18 BM were directly measured from six horses (dBM). Subsequently, the same horses were systematically photographed, and the identical set of 18 BM was measured indirectly from their images (iBM) by three operators, using an image analysis mobile app (ImageMeter®). Statistical analysis was conducted using JMP v16, with normality assessed through the Shapiro–Wilk test. Inter and intra assay coefficients of variation (CVs) were calculated between operators. A Bland–Altman test was applied to check the agreement between operators, and between dBM and iBM. The reproducibility of this technique was demonstrated by the mean inter and intra-assay CVs (6.4% and 7.9%, respectively), and by the agreement obtained between operators, which resulted in a bias of 0.8±0.8 cm and limits of agreement ranging from -2.7 to -2.4 cm. Moreover, a promising result was obtained regarding the precision of iBM when compared to dBM (bias of -3.9±1.3 cm, limits of agreement from -0.5 to -7.3 cm). The most accurate measurement was obtained with the ear's length, estimated with a precision of -0.2±0.3 cm (limits of agreement from 0.6 to -0.9 cm). Overall, the use of ImageMeter® represents a promising tool for researchers and breeders to estimate indirectly BM. Further studies on larger samples are needed to improve the methodology also considering the genetic implications

Phenotool: An information-sharing platform for livestock phenotyping methods and tools

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Animal phenotyping has undergone profound evolution in recent years with the development of high-throughput equipment, increasingly non-invasive sensors, the rise of "omics" technologies, and the use of artificial intelligence for image or sound analysis. The Phenotool platform has been developed with a twofold objective: 1) to present all the phenotyping equipment available within the LIPH4SAS research infrastructure; 2) to present the most important phenotyping tools available or under development for key traits related to the agroecological transition of livestock farming, mapped as part of a working group of the GIS Avenir Elevages, a French Think Tank dedicated to the future of livestock production. Phenotool consists of a web interface (www.phenotool.fr), developed under WordPress CMS, which allows online consultation of a database containing the main information available on phenotyping equipment. It offers an advanced search system allowing users to filter equipment according to the species investigated, the traits to be measured, or the geographical location. It also provides detailed information on each instrument listed, such as its description, technical specifications, and specific training materials for each instrument. Phenotool aims to facilitate access to information in the rapidly developing field of livestock phenotyping. By promoting the sharing of resources and knowledge, it should contribute to the emergence of new collaborations and innovative projects.

Feature identification for fattening pig body weight prediction: insights from correlation analysis

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Body weight is both a production parameter and a general health indicator for fattening pigs. Monitoring the body weight can therefore provide essential insights in pig production. Nowadays, pig body weight monitoring is either done by visual observation or by weighing devices on the group level. However, in order to go to individual pig health, welfare and productivity monitoring a more comprehensive understanding of body weight related parameters is required. In this study, the correlation of a range of feeding and drinking related features with the pig body weight was investigated. All features were calculated on the individual pig level for 4 fattening rounds of 60 pigs each (n=240 pigs) between 2018 and 2019, based on RFID, weight scale and flow meter sensor data. The correlation of pig body weight was largest with the following feed related features: average feed intake per visit in the last 24h (spearman correlation at optimal lag (r)=0.73 , lag=0), total feed intake in the last 24h (r=0.72 , optimal lag=-13), average duration of feeding visits in the last 24h (r=0.30 , lag=0), maximum duration of a feeding visit in the last 24h (r=0.16 , lag=-9) and feed intake in the last hour (r=0.11 , lag=-23). While correlation of pig growth (pig body weight increase) was largest, although small, with number of visits to the feeder in the last hour (r=0.19 , lag=0) , total feed intake in the last hour (r=0.16 , lag=0), total feed intake in the last 24h (r=0.06 , lag=0), average duration of visits in the last 24h (r=0.05 , lag=-1) and average feed intake per visit in the last 24h (r=0.04 , lag=0). These insight will be used further to develop a predictive model for pig body weight at the individual level.

Animal Science and Production in Italy

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The Italian Livestock industry is characterized by a marked regional distribution of species, breeds, and farming systems that reflects the wide geographical and climatic varieties of the Country, and socio-cultural contexts. Main livestock species farmed in Italy in 2023 (in millions of live animals) are cattle (5.4), sheep (5.9), goats (1.0), pigs (8.1), poultry (147.1). Other peculiar species are dairy buffaloes (0.44), rabbits (11.6), equine (0.48), and bees. In the Northern part of the Country specialized farming systems of pigs, poultry, dairy and beef cattle, can be found, together with the traditional semi-extensive alpine system based on local breeds. Dairy sheep and beef cattle are typical of the central Italy, together with a peculiar dairy buffalo industry. Small ruminants are the prevailing systems in Southern Italy. The gross income of the livestock industry represents the 29% of the agri-food industry. The livestock industry is supported by national breed Associations (AIA, FEDANA), agencies of the Government, organizations of farmers and producers' consortia. Italy owns a large number of PDO or PGI products (316 food products, 526 wines), most of which are of animal origin and that contribute to the 'Made in Italy' brand that is widely acknowledged all around the world. The structure of the Italian Animal Science research is mostly by Universities, the CREA of the Ministry of Agriculture, the CNR and regional Agencies of research. Most of Italian animal scientists are associated in the Italian Association for Animal Science and Production (ASPA).

Session 29

Theatre 2

Scientific perspectives on the sustainable development of our relationship with farm animals

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The scientific study of animal welfare emerged from societal concerns about the housing and management of farm animals. It is now a well-established discipline, encompassing fundamental investigations around the concept of 'animal welfare' and applied studies on welfare problems under commercial conditions. Assessing animal welfare has become a key area of scientific study, with the animal welfare indicators identified in such research increasingly included in quality assurance schemes. This trend is likely to continue, hopefully with greater harmonisation of indicators and a better use of the gathered data. However, focusing solely on monitoring the welfare of food-producing animals as a regulatory measure overlooks the broader context of our relationship with farmed animals. Our relationship is multifaceted and extends beyond the chain linking the animal to the farmer and the end consumer. Modern animal welfare science is embracing this complexity, as clearly demonstrated by the numerous interdisciplinary collaborations with nutritionists, geneticists, engineers, economists and others working within the broad area of animal production. Despite this, we still seem to be missing the bigger picture. One possible reason is that we are not looking back at the evolutionary history of these animals as a way to gain new scientific insights to help us in the future. Taking a scientific perspective, this presentation endeavours to reassesses our relationship with farm animals by considering their welfare within the framework of sustainable development. The interconnectedness of human, animal and environmental health is already highlighted in the One Health approach and the related One Welfare concept. The integration of animal welfare considerations with global challenges such as climate change, food security and antimicrobial resistance is increasingly recognised. Yet progress is painfully slow. A potential barrier seems to be that improved farm animal welfare is considered part of the problem, rather than part of the solution. By reassessing our relationship with farmed animals in a wider context, it becomes clearer that efforts to improve animal welfare can contribute even towards achieving the UN sustainable development goals.

Perspectives from the Animal Advocacy Movement

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Societal awareness of the plight of farmed animals and the ethical implications of their treatment is growing in the European Union, as shown by the success of the recent European Citizens' Initiatives (ECI) "End the Cage Age". This was one of three ECIs on animal welfare to gather more than 1 million signatures one after the other (the other two were "Save Cruelty-Free Cosmetics" and "Fur Free Europe"). End the Cage Age sent a strong political signal that the use of close confinement in animal production is no longer acceptable to the general public. Similarly, the Special Eurobarometer on Animal Welfare published in 2023 showed that more than eight in ten Europeans want farmed animals to be better protected. While market-driven animal welfare initiatives, such as the Better Chicken Commitment, can rapidly gain traction globally, policy-making is slow in addressing evolving ethical concerns. However, a heightened awareness of the needs of animals is reflected in the European Commission's proposal for stricter rules on the breeding and trading of dogs and cats, based on the five domains model. Although the Commission has delayed most of its legislative proposals on farmed animals, we anticipate that all future animal welfare legislation will be based on the five domains framework. To achieve a treatment of farmed animals that genuinely respects their complex needs, it will be necessary to reduce their numbers. This will imply rethinking our diets and how we produce food. The animal advocacy movement has joined a global call for a more ethical and sustainable food system that pays full regard to animal welfare, protects and regenerates the environment, and promotes human health and dignity.

Session 29

Theatre 4

Industry Insights

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Operators and representatives of the sectors linked to farmed animals (farmers, livestock traders, food industry, etc) state very often that the welfare of animals is a main interest for all the links of the production chain. Despite this statement would sound cliché, this sentence is completely true. Healthy well treated animals provide safe and high-quality products for human nutrition. It is a prerequisite for the sector represented by UECEV. While farmers have learnt and applied practices with the objective of providing safe products and doing huge efforts to meet societal expectations in terms of animal health and welfare, citizens and consumers seem to demand improvement of conditions of farmed animals even though their knowledge on animal farming is scarce. We need to look into the role of animal farming in society and vice versa, and to reflect on how to reconcile the views of the different parts of society regarding livestock activity and ensure that we have a broad vision of the objectives of livestock farming. Education is necessary to understand how food products arrive to our tables and how food of animal origin is obtained, and to understand that farming animals is a fundamental activity for our society. European primary educational programmes should include subjects on agriculture and farming, so the new generations know how to grow crops and how to obtain food from animals while respecting them and taking care of them. On the other hand, all the future progress in benefit of animals at all stages of their productive life must be done on sound basis of scientific knowledge.

The Consumer-Citizen Dilemma of Animal Welfare amid Farmer Protests and Agricultural Policy Failures
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Meat consumption sparks numerous controversies, leaving hardly anyone indifferent. Re-assessing our relationship with livestock is thus a pivotal task for both animal science and agricultural economics, as the sector's social license to operate is under scrutiny. Notably, contrary to long-term trends, meat consumption has recently declined in several countries, a phenomenon termed 'peak meat'. In Germany, meat consumption has decreased by approximately 10 kg per capita annually over the past decade. Initially, the animal husbandry, meat, and dairy industries grappled with animal welfare issues. However, there has been a notable increase in attention and willingness to adopt changes in recent years. Despite this shift, market success has been limited, prompting discussions on the limitations of a market-driven approach to farm animal welfare, commonly referred to as the "consumer-citizen gap". The issue is further complicated by the need to reduce animal husbandry volumes to meet environmental and climate protection goals. This dual pressure of declining sales, increased price competition, and substantial social transformation demands has created a challenging business environment. Given this context, the feasibility of driving such a transformation through market forces alone is increasingly questioned. In Germany, for instance, new concepts have been proposed, relying either on the governance of major food retailers or on comprehensive political regulation and funding to facilitate animal welfare improvements. However, both approaches have only been partially implemented and have faced significant resistance from farming communities. Drawing from his experience as the head of the German Commission for the Future of Agriculture, the author explores various options for a forward-thinking transformation policy that ensures the economic viability of livestock farming while addressing the pressing issues of animal welfare, environmental sustainability, and social acceptance.

Session 29

Theatre 6

Influence of early nutrition and nutritional programming on performance and physiological parameters of pigs

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Nutrition is one of the most important environmental influence and has a massive impact on the expression of the genetic architecture and thus the formation and function of the phenotype, not only acutely but also chronically. Especially in early life of pigs, i.e. the periparturient transition between the parenteral and the enteral route of nutrient supply but also around weaning when maternal milk is replaced by solid feed, are critical phases during which the animals are particularly sensitive to nutrition challenges. It is a concern that piglets born to hyperprolific sows have an increased risk of dying before weaning and might benefit from better nutrition. To ensure a healthy start in life that allows the utilisation of genetic growth and development potential as efficiently as possible, but also to promote animal welfare and reduce stress it is crucial to understand nutritional effects and their mechanisms in early life. It is important to know whether and how early nutritional effects carry over into later life, a concept known as early or nutritional programming, and whether this knowledge can be used to favourably influence pig health and performance. On this basis, nutritional strategies can be developed for dams and their offspring to improve the health and milk production of the mother on the one hand, and to promote early feed intake and improve the intestinal maturation of the offspring. Together with European partners, the Research Institute for Farm Animals Biology (FBN) in Dummerstorf, Germany, has contributed to this mission over the last 25 years. This review provides an overview of the relationship between pre- and early postnatal nutrition of pigs and the growth, physiological functions and health of the offspring and highlights some of the challenges for future pig farming. Funding: European Union; German Federal Ministry of Education and Research; German Federal Ministry of Food and Agriculture; German Research Foundation (DFG)

How to reduce enteric methane from dairy cows

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Enteric methane constitutes a predominant part of the carbon footprint of milk and the use of feed additives and manipulation of the nutrient composition of the diet are ways to reduce the emission of methane. The use of feed additives such as 3-nitrooxypropanol, nitrate and different types of seaweed have been shown to reduce enteric methane emission from dairy cows, but the concurrent effects on feed intake and milk production have in some cases been negative. Increased inclusion of fat in the diet reduces enteric methane emission primarily due to the dilution of the content of fermentable organic matter in the diet. Also increased inclusion of concentrate at the expense of forages high in fibre is a way to reduce the emission of enteric methane. Strategies related to changes in diet composition should always be accompanied by an evaluation of the effect on the carbon footprint of the diet.

A holistic approach for monitoring the environmental sustainability of the Italian Holstein cattle population R. Finocchiaro¹, L. Benzoni¹, G. Visentin², M. Dorigo³, F. Tiezzi⁴, M. Marusi¹, J. Layton¹, A. Bracchi⁵, G. Bonacina⁵, Z. Maddalena⁶, G. Gilson⁶, M. Cassandro^{1,7}

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A holistic approach for monitoring the environmental sustainability is implemented by ANAFIBJ. Since 2015 a wide range of environmental strategies to record data and to develop tools that meet community and farmer needs on mitigation climate change were developed. An ANAFIBJ pipeline was developed to incorporate new traits into the routine database. A consortium with various stakeholders for recording routine environmental traits for Holstein female population has been created. Methane emission records, milk-spectral records, ruminal content, and microbiota composition, collected from key individuals on the population, will feed into the central ANAFIBJ data flow system. Life Cycle Assessment (LCA) has been developed and inserted in a farmer tool for Holstein dairy herds. A “green passport” report was generated to summarize the innovative phenotypes records for each animal. Each tool plays a pivotal role in allowing farmers across the country to assess the environmental impact of their herd and inform decisions regarding herd management.

Measuring the Environmental Impact of a Local Cattle Breed via DHI Longitudinal Data

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The Agerolese cattle is a native breed of Southern Italy, primarily raised for milk production for the PDO cheese "Provolone del Monaco" (PMPDO). According to Taste Atlas, PMPDO is ranked as the 21st most renowned cheese globally, with a turnover of 3.7 million euros in 2022. The PDO region spans 163 km², encompassing mountains and coastal landscapes, where managing water, pastures, and anthropogenic pressures are current challenges. Animal diets must comply with the production regulation of PMPDO based on local pastures supplemented with GMO-free proteic fodder and hay. The current population consists of about 500 caws distributed in 30 herds. Besides the milk and cheese quality, the value of such a local breed is also represented by the ecosystemic services deployed, cultural heritage, and adaptation to the environment. Here, we quantify the environmental impact of the Agerolese breed, measuring the enteric methane emission based on Tier 2 DHI longitudinal data and IPCC 2019 equations. The average daily milk yield was 16.2 ± 3.4 kg d⁻¹, with 3.84 ± 0.29 and 3.39 ± 0.22 fat and protein percentage, respectively. The overall enteric methane emission averaged 0.03 ± 0.008 kg CH₄ kg FCM yield⁻¹, in line with other national or cosmopolite dairy breeds under more intensive selection programs.

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Theatre 4

Comparison of different residual carbon dioxide formulations for selection of feed efficient dairy cows

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Selecting dairy cattle for feed efficiency is a typical research area. Residual feed intake (RFI) has become the common metric for the evaluation of individual's feed utilization efficiency. A similar metric could be defined as residual carbon dioxide production (RCO₂). It has been found that expected feed intake, obtained by regressing dry matter intake (DMI) on energy sinks, may not be biologically plausible and this probably could be also the case for expected carbon dioxide production (CO₂). The objective of this study was to compare different RCO₂ and RFI formulations that are calculated using different partial regression coefficients for energy sinks which are obtained either from regression on energy sinks or from different energy requirement standards. Repeated daily GreenFeed measurements of CO₂ (n = 49155) and DMI records from 82 primiparous Nordic Red dairy cows were used. Three types of RCO₂ and RFI formulations were calculated. The first was by fitting a multiple linear regression (RCO₂MLR and RFIMLR) whereas the second and third were extracted from Finnish energy requirement formulation (RCO₂FIN and RFIFIN) and National Research Council 2021 (RCO₂NRC and RFIN-RC), respectively. Correlations between different RCO₂ and RFI formulations ranged from 0.37 to 0.46, implying that different formulations lead to selection of different animals. Correlation between RCO₂ and RFI was highest when the formulations were based on the Finnish energy requirement formulation and lowest when formulations were based on the estimated partial regression coefficients. Selection based on RCO₂ formulations would lead to improvement in energy conversion efficiency albeit with slightly lower rate compared to selection based on RFI formulations.

Circadian variation and animal-wise association between methane and a new feed efficiency trait in Nordic Red dairy cows

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Animal-based methane (CH₄) mitigation via improved feed utilization efficiency (FE) requires identification of accurate FE metrics and estimation of its associations with other traits. One potential FE metric in dairy cows is residual carbon dioxide (RCO₂) but its associations with CH₄ output traits is unclear. Besides, currently most averages of CH₄ are estimated from spot-samples of 3 to 30 min over a period of time. This may affect prediction of CH₄ and understanding the circadian variability is essential. The objectives were to assess the circadian variability of CH₄ emission and estimate the association of CH₄ phenotypes with FE metrics. Data was from 137 primiparous Nordic Red dairy cows of Luke's Jokioinen research farm and included 51,977 repeated records. CH₄ and CO₂ were measured using 2 Greenfeed devices. Data was edited and CH₄ production, intensity and yield were derived. The FE metric RCO₂ was defined as a difference between actual and predicted CO₂ output. Circadian variation of CH₄ was estimated by fitting a model containing time of measurement as a fixed effect. Animal correlations were calculated. The correlations between CH₄ phenotypes and RCO₂ ranged from 0.51 to 0.69 and ranged from 0.21 to 0.33 with residual feed intake. Significant circadian variability with five marked elevations of CH₄ was observed: one in the morning and four in the afternoon and late evening. The high and positive animal correlations between RCO₂ and CH₄ output traits indicate the potential of RCO₂ metric for improving FE whilst reducing CH₄ emission. The high circadian variability indicate caution in quantifying CH₄ emission for a given period based on too sparsely taken spot samples.

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Theatre 6

Correlation between mid-infrared-based predicted methane production and milk urea in Walloon Holstein dairy cows

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Nitrogen (N) losses to the environment and greenhouse gas emissions (GHG) as methane (CH₄) and nitrous oxide are two of the main environmental concerns of dairy cattle farming. The low level of N utilization of dairy cows results in approximately 60% of ingested N being excreted as urinary urea (UU). Although UU is difficult to measure, milk urea (MU), a correlated trait with UU, can be measured at a low cost and is included in most milk recording systems. Accurate measurements of CH₄ production require complex and expensive techniques; however, milk mid-infrared (MIR) data have been proved as a fast and cheap method for predicting daily CH₄ produced by individual cows. Genetic selection is a useful tool for reducing N and CH₄ emissions in dairy cattle but requires estimating the genetic correlation between the trait of interest and the traits present in the current breeding goals. This study aims to estimate genetic parameters for MU and CH₄ in Walloon Holstein cows. The data was collected from 2006 to 2022 on 229,465 first-parity Holstein cows (1,529,282 test-day records) distributed in 1,530 herds. The MIR-based predicted methane emission (PME, g/d) and predicted methane emission intensity (PMI; PME/ kg of milk) were used. The PMI was then log-transformed to be normally distributed (LMI). Random regression test-day models were used to estimate genetic parameters through the Bayesian Gibbs sampling method. The means (standard deviation (SD)) of PME, LMI, and MU were 324.3 (68.20) g/d, 2.67 (0.38), and 25.38 (8.02) mg/dl, respectively. Mean heritability estimates for PME, LMI, and MU were 0.13 (0.04), 0.25 (0.05), and 0.21 (0.02), respectively. Genetic correlations estimated between PME, LMI, and MU with milk yield, fat and protein percentages (MY, FP, PP) ranged from -0.12 (MY) to 0.42 (FP), -0.89 (MY) to 0.57 (FP), and 0.01 (PP) to 0.05 (MY), respectively. The mean genetic correlations estimated between MU and PME (-0.02; SD = 0.05) and LMI (-0.03; SD = 0.06) were close to zero indicating no association between these traits. The results demonstrated large variations for PME, LMI, and MU encourage the implementation of selection for environmentally friendly cows.

Estimating Direct and Maternal Effects on Residual Metabolizable Energy Intake in Holstein Calves

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The two the largest expenses in the dairy industry are the animals' feed and the rearing of heifers. While in many countries feed efficiency in lactating cows has already been integrated into the genetic evaluation, studies in dairy calves are still scarce. Because maternal effects are known to influence important traits measured early in life, they may play an important role in dairy calf feed efficiency. The objective of this study was to estimate genetic parameters of feed efficiency in pre-weaned dairy calves and investigate the significance of maternal genetic effects on feed efficiency. Residual metabolizable energy intake (RMEI) of 471 Canadian Holstein calves in two time periods (RMEI1: first month of age; RMEI2: second month of age) was used as a measure for feed efficiency. Statistical analysis using animal models including maternal effects was performed with ASReml. Maternal effects significantly (p -value= 0.04) improved model fitting of RMEI1, with high negative genetic correlations between direct and maternal effects (-0.88 ± 0.02). Without considering maternal effects, heritability estimates for RMEI1 and RMEI2 were 0.19 ± 0.11 and 0.32 ± 0.12 , respectively. RMEI1 the direct heritability was 0.15 ± 0.13 , the maternal heritability was 0.27 ± 0.12 and a total heritability of 0.02. The estimated genetic correlation between RMEI1 and RMEI2 (0.77 ± 0.36) indicated that RMEI in the two periods may be considered separate traits. Further studies with more animals and herds, as well as an investigation on the potential relationships with other important traits should be carried out to understand whether and how RMEI could be incorporated in selection decisions. Despite the limited dataset used in this study, moderate heritability estimates indicate that selection for more feed efficient calves is possible.

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Theatre 8

Genetic relationships between methane emissions, dry matter intake, bodyweight, and milk yield of dairy cows

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Animal breeding is recognized as an effective strategy to mitigate methane (CH₄) emissions from dairy farming. However, CH₄ is challenging to record and genetic correlation estimates between CH₄ and other breeding goal traits are scarce, while they are essential to construct balanced selection indices and to pinpoint possible indicator traits. The aim of this study was to estimate genetic relationships between CH₄ emissions, dry matter intake (DMI), bodyweight (BW), and milk yield traits in Dutch dairy cows. Enteric CH₄ emissions were recorded with sniffers in the feeding bins of milking robots on 7,749 cows from 72 farms. Data on DMI (3,526), BW (3,783), and milk traits (3,879) were available on cows measured at WUR farms. Genetic parameters were estimated, using bivariate restricted maximum likelihood models with an H-1 relationship matrix in ASReml. The genetic correlation between CH₄ and DMI was moderate and was 0.21 ± 0.12 , and the correlation between CH₄ and BW was low (0.06 ± 0.11). Genetic correlations between CH₄ and milk production traits were positive, and were: 0.25 ± 0.12 for milk yield, 0.07 ± 0.11 for protein percentage, and 0.18 ± 0.10 for fat percentage. The preliminary results show that there are moderate undesired correlations between CH₄ and production traits, which should be considered in future breeding programs.

The potential of residual feed and energy intake as direct selection traits for Fleckvieh cows

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In order to select for higher feed efficiency, direct and indirect traits need to be defined, which can be implemented into existing breeding programs. Direct traits can either be residual or ratio traits, with residual traits considered more suitable for genetic evaluation. The aim of this study was to develop a model for residual feed (RFI) and energy intake (REI) as direct efficiency traits for Austrian Fleckvieh cows (dual-purpose Simmental). Furthermore, RFI and REI were compared for their suitability to describe efficiency. Performance data of the research farm of the Federal Agricultural Research Centre in Raumberg-Gumpenstein were used to develop models for RFI and REI. A total of 19 Fleckvieh cows with overall 68 lactations were included in the analyses. The final models used for comparison included the most important energy sinks (i.e., milk production, maintenance, body weight change). Their predictive power for the actual feed or energy intake was moderate ($R^2 = 0.50-0.60$). When comparing RFI and REI models, the REI models had a slightly better fit. Larger data sets are needed, however, to further validate the models and to decide if RFI or REI is better suited for genetic evaluation.

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Theatre 10

Genetic correlation between body weight and methane emission in Norwegian Red

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Body weight of dairy cows is of particular interest as selection for increased milk production leads to larger animals which comes with a cost. Increased automatization of herds has enabled frequent recording of body weight in lactating cows. Our aim was to analyse body weight (BW) in Norwegian Red cows genetically, and to estimate genetic correlation to daily methane (CH₄) emissions. Data was available from commercial herds with weight scale for BW and GreenFeed for CH₄ measures installed. Records from 2019 to 2024 were available from 25 farms with GreenFeed and among these 9 herds contributed with data on BW for lactating cows. Quality control and data edits left us with 260 132 and 220 932 records on daily BW and CH₄ from 1 960 cows, respectively. 410 cows had records on both traits. Traits investigated were average daily BW (Kg) and average daily CH₄ (gram per day). We used a bivariate linear animal repeatability model with common herd testday, permanent environment, and additive genetic effect of animal as random effects. The (co)variances were estimated using the DMU package. Heritability (standard error) of BW and CH₄ was 0.58 (0.05) and 0.39 (0.04), respectively. The estimated genetic correlation between the traits was 0.50 (0.09). Our results confirms that body weight is a highly heritable trait, and there is an unfavorable genetic correlation between BW and daily methane production in Norwegian Red, thus increased body weight would also increase level of methane.

Describing feed efficiency using metabolomic data in pigs

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Feed conversion ratio (FCR) is commonly used to determine production efficiency in pigs. However, the basic biological mechanisms of this complex trait are still largely unexplored. In this study, we aimed to describe FCR in Italian Large White pigs using metabolomic data. For 691 pigs, FCR random residuals (FCR-RR), untargeted plasma metabolomic data (constituted by about 1000 metabolites) and high-density single nucleotide polymorphisms were available. We first identified two extreme groups of pigs (100 with the lowest and 100 the highest FCR-RR) and compared their metabolomic data using Sparse Partial Least Squares Discriminant Analysis and Boruta algorithm. With the combination of these two approaches, we identified 12 metabolites that could discriminate the two extreme groups of pigs. These metabolites were involved in a few metabolomic pathways, including fatty acid oxidation, carnitine, alanine, and histidine metabolism. Genomic heritability of the selected metabolites ranged from 0.11 to 0.35. The obtained results can contribute to identify molecular proxies useful to describe feed efficiency and develop new breeding strategies in pigs. Acknowledgments: This study has received funding from the European Union – NextGenerationEU under the National Recovery and Resilience Plan (PNRR) – FEEDTHEPIG, proposal code P2022FZMJ9 – CUP J53D23018310001 and from the European Union’s Horizon Europe research and innovation programme under the grant agreement No. 01059609 (Re-Livestock).

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Poster 12

Network analysis unraveled the complex interactions in the rumen microbiome associated with methane emission in dairy cattle

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Methane emissions from ruminants, particularly dairy cattle, represent a significant source of greenhouse gas (GHG), contributing to climate change. This study employed Weighted Gene Co-expression Network Analysis (WGCNA) to investigate the complex interactions within the rumen microbiome that influence methane emissions. By integrating rumen microbiome sequencing data with methane emissions measurements from 750 Holstein cattle, we employed a quality-based bioinformatics pipeline to identify microbial communities and their associations with methane production. Meanwhile, our findings highlight the crucial role of both archaeal and bacterial communities in methane emissions, revealing specific microbial modules (MEblue, MEyellow, METurquoise, and MEBrown), interactions (Clostridium–Clostridium–Prevotella-RF39–Clostridium) and different taxa (archaeal genera VadinCA11, Methanobrevibacter, and Methanosphaera; bacterial taxa Bacteroidales, Ruminococcaceae, Prevotella, Treponema, Clostridium, Anaeroplasma, CF231, and Succinogenes) significantly correlated with methane emissions. Methanobrevibacter, and Methanosphaera, archaeal phylum Euryarchaeota, are known for their methanogenic capabilities and predominant abundance for archaeal communities. In addition, bacterial genera like Prevotella, and Clostridium can produce volatile fatty acids (VFAs) and hydrogen, which can be substrates for methanogenesis in archaeal communities. The application of network analysis provided a systematic understanding of the microbiome-methane emission relationship, providing its potential as an innovative approach to study microbiome-trait association in cattle.

Investigating microbiability and microbiome-wide association analysis across diverse gut sections in growing-finishing pigs fed nitrogen- and phosphorus-restricted diets

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The reduction of nitrogen and phosphorus (N/P)-excretion can be achieved by improved feeding strategies and/or by breeding more nutrient efficient pigs. In this context, our project aims to quantify the effects of N/P-restricted feeding systems and to estimate the heritability (h^2) and microbiability (m^2) effects on feed N/P-related traits. One open question is the role of different gut sections (jejunum, ileum, caecum, and colon) related to the composition of the microbiome and its impact on different efficiency traits. In total, 208 samples of 4 gut sections (jejunum, ileum, caecum, and colon) obtained from 52 crossbred pigs were collected and amplicon sequencing (16S rRNA) was performed. Significant differences for microbial families and genera were observed between small and large intestine in terms of alpha and beta diversity. In general, the estimates for h^2 were higher compared to the estimated of m^2 . The microbiome of small intestine influenced the expression of N/P-digestibility, while high m^2 values for two feed efficiency traits were observed in large intestine. For nearly all other traits, m^2 values were zero. Additionally, a microbiome-wide association study revealed a high number of gut specific amplicon-sequence variants (ASVs), with very small effects on 5 efficiency traits. This might be due to the small number of observations. In conclusion, the results of this study revealed gut specific microbial variance affecting efficiency traits. The study was supported by funds of the MULNV (project-no. 41.2019.03) and of the LANUV (project-no. 17-02.04.01-08/2019), North Rhine-Westphalia, Germany.

Session 30

Poster 14

Genetic architecture of feed efficiency traits in American mink

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Optimizing feed efficiency (FE) in American mink is crucial, as it holds the key to substantial cost savings within mink production systems. The study aimed to estimate genetic parameters for various FE and their component traits, while also revealing the genetic background underlying these key traits. Phenotypic data from 2,288 American mink in two Canadian farms were recorded with genotypes obtained using an Affymetrix 70K single nucleotide polymorphism (SNP) array. In addition, the characteristics of copy number variations (CNVs) and runs of homozygosity (ROH) islands within the genome of American mink were identified by analyzing the whole-genome sequencing data of 100 individuals. Moderate heritabilities and favorable genetic correlations among FE traits in American mink support their inclusion in genetic/genomic selection. A total of 34,652 ROH segments were identified across all individuals. Predominantly, shorter segments (0.3–1 Mb) constituted approximately 84.4% of all ROH throughout the genome. Within these segments, 63 ROH islands were identified. Identifying a total of 164,733 CNVs consisting of 144,517 deletions and 20,216 duplications, the study merged overlapping CNVs to form 5,378 CNV regions (CNVRs). This collectively covered 47.3 Mb (1.9%) of the mink autosomal genome. Through association analyses using the mixed linear model, 42 SNPs were found to have significant associations with six FE traits. Enrichment analyses of candidate genes overlapped with identified CNVRs, ROH, and significant SNPs revealed several pathways related to growth, lipid metabolism, and immune response. Overall, this study provides valuable insights into the genetic architecture of FE traits and identifies potential candidate genes associated with these complex traits, which can be implemented into breeding programs.

Effect of paternal breeding values for residual feed intake on reproductive performance of Hereford heifers
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Feed efficiency (FE) is a key trait for animal production. Residual feed intake (RFI) is the most explored FE trait due to its moderate heritability and independence of animal performance. Estimated Breeding Values (EBVs) for FE in Hereford are available in Uruguay, and a comprehensive investigation of the associations of FE with other traits, including reproductive performance is ongoing. We investigated some reproductive traits of 347 Hereford heifers born in springs of 2017 to 2020 from sires of high and low EBVs for RFI and raised in grazing conditions. Animals were classified into two groups according to their paternal FE-EBV. These EBVs are based on RFI but expressed as feed saved, implying that lower RFI leads to large feed savings (high FE, H, 12 sires, mean EBV=107, range:103-115; n=212, and low FE, L, 7 sires, mean EBV=93, range:82-98). At 25m of age, and previous an estrus synchronization with Prostaglandin, the ovarian cyclicity (OC) was assessed by ultrasonography, and the presence of corpus luteum was registered. Artificial insemination (AI) was performed twice a day 12h after estrus detection for 15d. Heifers were exposed to bulls for 70d on a 1:45 ratio, 25d after the beginning of AI. Pregnancy was assessed 45d after the end of the breeding season. The OC and pregnancy frequencies of H and L heifers were compared with GLIMMIX procedure of SAS using a binomial distribution. The statistic model included EBV group and year as fixed effects. Paternal FE-EBV did not significantly affect neither OC (H 59% vs L 47%, P=0.0894) nor pregnancy (H 92% vs L 99%, P=0.9247). These results indicate non unfavorable impact of improving FE on heifer's reproductive performance, although additional traits are being studied.

Deep phenotyping of feed efficiency in individual dairy cattle using longitudinal and average feed quality data

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Feed efficiency of dairy cows is a trait affected by multiple components. These inner, invisible components of individual cows are named deep phenotypes. Deep phenotypes were assessed in a pilot study using the mechanistic model LiGAPS-Dairy and a genetic algorithm. The pilot study, however, used average feed quality (FQ) data at herd level, which is expected to reduce the accuracy of deep phenotypes. Therefore, the objective was to investigate whether deep phenotyping improved by using individual, longitudinal FQ data compared to average herd FQ data, which was tested in two datasets: TGEN and LELY. From each dataset, multiparous Holstein cows (n =110) with ≥ 5 weekly records for FQ (digestibility and DVE: intestinal degradable protein) and ≥ 10 weekly records for the measured phenotypes dry matter intake, liveweight, and milk yield were selected. Deep phenotype accuracy was reflected by the sum of errors between simulated and measured phenotypes. For all cows, the model was run with FQ from the pilot project (RUN1), the average herd FQ of the dataset (RUN2), the average FQ per individual cow in the dataset (RUN3), and individual, longitudinal FQ data with interpolated missing FQ values (RUN4). The sum of errors improved considerably for TGEN data moving from RUN1 to RUN4: -147.40, -72.23, -70.93, -53.87. For LELY less improvement was seen from RUN2 to RUN4 (-97.30, -98.79, -95.83). Also, RUN1 (-88.45) gave unexpectedly the best fit using LELY data. Whether the better model fit for TGEN might stem from the estimated DVE values or the frequency of FQ recordings is being investigated. In conclusion, using individual longitudinal records improved deep phenotyping compared to herd or cow's averages.

Cosmopolitan and local Italian beef cattle breeds uncover common patterns of heterozygosity related to fitness traits

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The investigation of heterozygosity is less common compared to the study of homozygous patterns. However, Heterozygous Rich Regions may harbor significant loci for immune response, survival rate, and fertility traits. For this reason, the heterozygosity patterns characterization was conducted in five beef cattle breeds, which included two cosmopolitan breeds (Limousine and Charolais) and three local breeds (Sarda, Sardo Bruna, and Calvana). A high degree of heterozygosity using a consecutive runs approach was identified. Several regions exhibited recurrent heterozygosity peaks and were consistently found on specific chromosomes across all breeds, specifically autosomes 15, 16, 20, and 23. The cosmopolitan, Sardo Bruna, and Calvana breeds also displayed peaks on autosomes 2 and 21. Forty-two top runs shared by more than 25% of the population were identified. These genomic fragments encompassed 18 genes, two of which are directly linked to male fertility, four are associated with lactation, two genes play roles in survival and immune response, and a region related to growth and carcass traits was detected in Limousine. The study revealed particular segments of the cattle genome linked to various functional traits. Unexpectedly, heterozygous regions are common across cosmopolitan and local breeds. The genes identified have potential applications in breeding programs and conservation studies to investigate the phenotypes associated with these heterozygous genotypes.

Estimating effective population size from genomic rates of coancestry and inbreeding

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Effective population size (N_e) is a key parameter in quantitative genetics and evolutionary biology. Commonly, estimates of N_e are derived from rates of coancestry and inbreeding. Although traditionally these rates have been obtained from pedigree data, they can also be obtained from genomic data in order to increase the accuracy of the estimates. Using stochastic simulations, this study aims at assessing N_e estimates derived from a range of genomic measures of coancestry (f) and inbreeding (F) by comparing them with true identity-by-descent (IBD) values. Genomic measures included those described by Li and Horvitz (fLH1, FLH1), VanRaden (fVR1, FVR1, fVR2, FVR2) and Yang and colleagues (fYA1, FYA1, fYA2, FYA2). Three management strategies – Random Contribution (RC), Equalized Contribution (EC) and Optimal Contribution (OC) – were considered. For all strategies, N_e estimates derived from f were closer to IBD values than those derived from F . Additionally, N_e estimates based on f measures were more stable and accurate than those based on F measures, as coefficients of variation across replicates were lower and correlations with the true values were higher. Under strategies RC and EC, all f measures led to N_e estimates close to the true values. The highest correlations between N_e estimates with the true values were observed when using fYA2 and fVR2, and the lowest when using fLH1. The optimization of contributions under OC, led to a N_e lower than the true value, particularly when using fLH1. Also, correlations with the true values were very low for any estimator. The study highlights the importance of choosing appropriate estimators for evaluating the evolution of N_e when managing breeding programs.

Genomic analysis of diversity and population structure of baroque horse breeds in Czech Republic and Slovakia

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The Old Kladruber and the Lipizzaner are two old Baroque horses adapted to Central European conditions with a common history. The origin of both breeds goes back to the 16th century. In the course of the common history of these two breeds, a certain exchange of genetic material took place, mainly between these two breeds, but also with other horse breeds. Our main objective in this analysis was to assess the conservation status, population structure and admixture of the Old Kladruber Horse (OKCZ) and Lipizzaner (LIPCZSK) breeds in the Czech Republic and Slovakia. We also estimated the genetic relatedness of these breeds with other Baroque horses and neighbouring horses. The analysis included 233 horses of the OKCZ breed (the subpopulations with grey and black coats), 73 horses of the LIPCZSK breed and 230 horses of other breeds (mainly warmblood breeds). The analysis was based on 51,166 autosomal SNPs genotyped with the GGP Equine 70k chip. In general, the observed genetic diversity (heterozygosity and haplotype richness) was close to the values observed in other breeds. On the other hand, genomic inbreeding coefficients (FIS and ROH-based inbreeding) were relatively high. While the estimates of the current effective population size of the Old Kladruber horse were 48 individuals (for the grey and black varieties) and Lipizzaner were 67 individuals, the estimates of the historical effective population size showed a significant decrease ten generations ago. The estimated contemporary effective population size demonstrates the need for careful monitoring of genetic diversity. Our results will thus contribute to breeding and conservation strategies for these breeds. This study was supported by the projects GACR 24-14325L and APVV-20-0161.

Identifying genetic loci influencing body weight in the endangered dual-purpose German Black Pied (DSN) cattle breed

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The German Black Pied (DSN, “Deutsches Schwarzbuntes Niederungsgrind”) is an endangered dual-purpose breed counting approximately 2,500 DSN herdbook cows. Previous genetic studies on DSN have largely focused on milk production traits and disease resistance. This study focused on growth traits within the DSN breed with measurements at different growth stages: at birth and at the average age of 3 weeks, 2 months, and 1.5 years. Employing imputed whole-genome sequencing data of 565 DSN bulls and linear mixed models, we identified five loci associated with birth weight on BTA 2, 5, 8, 12, and 18 and one locus associated with body weight at the average age of 3 weeks on BTA 11. Interestingly, the locus on BTA 5 for birth weight was in the vicinity of the gene *MGST1* locus which we identified previously for milk fat content in DSN cows. For the other identified loci, we found no overlap with loci associated with milk performance traits. This suggests that in dual-purpose DSN cattle, genes with a significant impact on growth or milk production are largely unlinked, meaning they are inherited independently. Our research not only identified unique genetic loci associated with growth but also explored the interplay between growth and milk production. These insights offer a foundation for developing a conservation and breeding strategy that maintains the DSN breed’s dual-purpose character, ensuring its long-term viability and utility in both dairy and meat production.

The benefits of genomics in small populations: case study of type traits in the Dual-Purpose Blue cattle breed
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The Dual-Purpose Blue (DPB) is a small cross-border breed from Belgium and the North-East of France. This local breed was preserved by breeders for its robustness and ease to handle. Several transboundary projects have resulted in the development of common genetic evaluations for this breed, including the estimation of (genomically enhanced) estimated breeding values ((G)EBV) for linear type traits. Once in a year, EBV and associated reliabilities (REL) are obtained using a multiple-trait animal model and then, subsequently used in a pseudo single-step GBLUP with Bayesian integration to obtain GEBV and associated genomically enhanced REL (GREL). This study aimed to assess the gain in REL for type traits of DPB when genomic information is added. Phenotypes included 23 traits, 18 individual and 5 overall traits (udder, development, feet and legs, muscularity, rump). All traits were scored from 1 to 9, except for stature, which was measured. In total, 10,360 records were used, with 2,195 animals being genotyped. Pearson correlations were computed between EBV/GEBV, and REL/GREL. Resulting correlations between EBV/GEBV, and REL/GREL were very high for all type traits (0.96-0.99). In general, REL enhancement for genotyped animals was observed, ranging on average from 0.07 to 0.13 points across traits, which is in line with our expectations. Specifically for genotyped bulls, REL enhancement ranged on average from 0.11 to 0.14 (maximum from 0.29 to 0.35) across traits. This clearly indicates the benefits of the use of genomic data, even in a small population, which will allow better selection decisions.

Session 31

Theatre 6

Genetic analysis of calf survival in a critically endangered antelope (Nanger dama mhorh)

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Mhorh gazelle (Nanger dama mhorh) is a Sahelo-Saharan antelope that is currently extinct in the wild. Its captive breeding program provides an important tool for rearing sustained populations, with offspring survival playing a critical role. The aim of this study was to determine the genetic parameters of perinatal (PS) and weaning survival (WS) of mhorh gazelle calves, and to investigate the possibility of using these fitness traits as breeding objectives in the conservation program. Up to 2185 calf survival records from the studbook were used in the analysis. All genetic parameters were estimated via mixed linear models within a Bayesian frame and using the TM program. The predictive value of the different models was compared using the logCPO value. The calf model had the best fit with data for the PS and the calf-permanent model for the WS. Heritabilities estimated for the direct genetic effect were moderate, 0.279 for PS and 0.157 for WS. The maternal environmental component in the WS was 0.048. Calves of multiparous dams had a higher PS, but there were no significant differences in WS depending on the number of calving. For both traits, male calves had a lower probability of survival than females, and offspring born from young and old gazelles were less likely to survive. High inbreeding coefficients of the calf reduce PS and high inbreeding of the dam make descend WS. Neither PS nor WS were affected by the maternal genetic effect, so mating strategies in the conservation program should focus on the direct genetic effect to increase the potential of the population to respond to selection on offspring survival rather than on the maternal contribution.

Towards a generic measure of genetic relatedness of populations

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The extent to which two breeds are genetically one population is important for deciding whether endangered breeds should be managed and conserved separately. Examples are transboundary breeds, different varieties within a breed or endangered breeds with a regular influx of breeding stock from another breed. Real-life cases vary in population size, pedigree depth, history and exchange of breeding stock. Our objective is to develop a generic measure of genetic relatedness between populations to support decision making in conservation of endangered breeds. One of the methods is the average relatedness between breeds (a_{ij}) and within breeds (a_{ii} and a_{jj}). The kinship distance ($a_{ii} + a_{jj} - 2a_{ij}$) is directly related to the drift variance ($(a_{ii} + a_{jj} - 2a_{ij}) * \text{VarG}$) of traits. It indicates how much a trait drifts in the same direction in two related populations due to random genetic drift. Genetic drift variance between related populations (GDVB) is lower than between unrelated populations. We propose to use retained GDVB as an objective measure of uniqueness of populations. It is calculated as the estimated GDVB as a fraction of the GDVB if these populations were unrelated. Between Dutch and Belgian draught horse populations, retained GDVB gradually reduced to 55% between 1970 and 2020. The proposed measure can be estimated from pedigree, genomic information or both. If this is not available, the development of retained GDVB over time can be approximated with simulation using the details of both breeding populations. A generic measure allows national coordinators for animal genetic resources to use more objective criteria regarding the uniqueness of breeds.

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Theatre 8

Derogations in breeding programmes for endangered breeds according to EU animal breeding legislation (EU 2016/1012)

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The main tasks of the European Union Reference Centre for Endangered Animal Breeds (EURC-EAB) include working on harmonised methods for the preservation of endangered livestock breeds and informing breed societies and competent authorities on those matters. An important instrument for the facilitation of breeding those breeds are the specific derogations in breeding programmes for endangered animal breeds and hardy sheep breeds, of which breeding organisations can take advantage of according to (EU) 2016/1012. The survey conducted by the EURC-EAB showed, that 66% of the participating breed societies with breeding programmes for endangered breeds have already implemented such derogations. The most frequently used derogations are the upgrading of male progeny to the main section of the herd book, followed by restricting breeding with purebred breeding animals and implementing lower criteria for performance recording and genetic evaluation in comparison to mainstream breeds. The vast majority (87%) of the corresponding breed societies described the derogations as helpful and 65% stated the process of implementation as easy. Nevertheless, 16% of the breed societies reported about specific problems in the implementation process. The presentation will give a detailed view on the survey results concerning the derogations and concrete examples of different types of derogations used by breed societies from EU Member States.

Simulation of genetic rescue from a gene bank using a complex load structure

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The main focus of conservation breeding and especially gene-banking is to safeguard genomic variation of farm animals. Genetic rescue with semen from a gene bank restores lost variation, but holds the risk to reintroduce already purged deleterious variation from a formerly large population into a then endangered population with consequently low effective population size. We modeled six scenarios of populations that start with an initial size of 1000 individuals and are moderately selected for a performance trait for 60 generations. All populations experience a severe decrease in population size (from 1000 to 800, 600, 400, 200 and 50, respectively) between generation 18 and 26. The population is rescued in generation 61 with gene bank semen from before and after the decline, and randomly mated for another 30 generations. Deleterious load was modeled as a composite of homozygously lethal and moderate variants. The first generations showed a severe decrease of litter size due to deleterious load which was alleviated by purging before the populations went into the size bottleneck. Directly after, the smaller scenarios litter size dropped again, before it leveled around generation 60. Purging was more efficient in smaller populations, removing nearly all lethal alleles, while a basic load, made up from low-effect alleles persisted. Rescue with gene bank animals led to a reduction in litter size in all cases. Litter size depression in scenarios 200 and 50 was stronger than the initially modeled depression of about 12 %. We found that this was also due to reintroduced lethal alleles. Moderately deleterious mutations went more often into fixation after rescue, as selection against them was less effective than in the more diverse population in the beginning, and contributed to a basic load compromising litter size permanently. Especially in the smallest scenario rescue with animals from generation 26 carried about 50 % of load in fixed alleles.

Session 31

Theatre 10

Sheep along the trading routes: what may we tell from their demographic history? A case study from Hungary

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Sheep (*Ovis aries*) has played a major role in farming for food, textiles, and clothes since their domestication (ca. 11,000 years ago). Subjected to intense transportation, sheep have a complex demographic history. We examine genetic diversity and historical demography of sheep breeds from Hungary and neighbouring areas. We analysed a total of 763 animals from 39 breeds, using genotyping data from the SMARTER database. Genetic diversity, population structure and individual genetic components analyses revealed a clear split in two subpopulations for the Hungarian Racka breed, maybe related to the different colour variants, and a low level of admixture for both Hungarian Racka and Tsigai. Several breeds showed introgression from Merino or Texel and gene flow between domestic breeds and their wild relatives. Historic trends in effective population size and runs of homozygosity revealed recent bottlenecks, confirming previous results and in agreement with documented demographic changes. Overall, our results provided clues on the possible events related to their early arrival in this area: the inferred admixture events, spanning from the Bronze Age to early Middle Ages, suggested bi-directional gene flow in several waves of contact.

First insights in the genetic structure of Cyprus's indigenous cattle

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Indigenous Cyprus cattle significantly influenced local agricultural development, dietary changes, and cultural heritage. Two native morphotypes, exhibiting all zebu characters, were reported from well-defined geographic regions of the island. The “Mesaoria” type occurs in the central-eastern part of the island, with a larger body size and a thinner coat compared to the “Paphos” type occurring in the south-western part. Considering the Cypriot, diverging from the primary production, economic model as well as the low growth rate and milk productivity compared to genetically improved breeds, the local population was until recently threatened with extinction as the total number of individuals from the 1960s to 2008 decreased by 97%. Nevertheless, thanks to the joint efforts of committed cattle breeders and governmental subsidy programs, this hardy and well-adapted breed has maintained a constant population size of ~1200 individuals over the last decade. Herein, we aimed to conduct a comprehensive evaluation of the local breed, whose results would lead to the genetic characterization and hence promote ongoing and future conservation efforts. For this purpose, genome-wide SNPs were retrieved using the Illumina Bovine 777K array from 96 individuals sampled across the island. Population genetic metrics examine the genetic divergence of the native types as well as the genetic structure of the sampled population and their relationship with commercial, genetically improved cattle breeds on the island. Beyond conservation of biodiversity, ecosystem services, and sustainable agriculture, the preservation of this breed is essential for Cyprus's cultural heritage.

Session 31

Theatre 12

Exploring genomic selection opportunities in Italian local cattle breed

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Here we assessed the impact of genomic selection (GS) on 4 Italian local cattle breeds: Rendena (REN), Reggiana, Alpine Grey, and Valdostana. Our objectives were i) to compare current models (PBLUP) with GS for milk yield, and ii) to evaluate potential gains due to GS. To integrate SNP data with production records, we included phenotypic data >2010. Imputation achieved an average accuracy of 95%, and genomic EBVs (gEBV) were estimated with ssGBLUP. PBLUP and ssGBLUP were compared using a LR approach, with young animals serving as validation cohort. Across all breeds, ssGBLUP consistently outperformed (i.e., in REN 0.31 vs. 0.45 of accuracy for PBLUP and ssGBLUP, resp.). Notably, ssGBLUP showed higher accuracy in gEBVs, particularly in young bulls. Comparing current progeny testing to scenarios involving selection of young bulls on gEBVs, we observed greater genetic gains with GS, along with a reduction in inbreeding due to a larger pool of reproducers. This study highlights the favourable application of GS in local cattle breeds. Acknowledgements: Funded within the “Agritech” – Next-Generation EU PNRR – Missione 4 Comp. 2, Invest. 1.4 – D.D. 1032 17/06/2022, CN00000022. Also funded within PSRN – Dual Breeding 2 (CUP J61J18000030005, J51J18000000005, J71J18000020005, J81J18000030005), and by PRIN 2022 – Research Project n. 2022F43HWL.

Genome-wide approaches reveal novel genetic markers potentially involved with Visna Maedi virus infection in Valle del Belice sheep

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Visna/Maedi virus (VMV) is a complex production-limiting ovine lentiviral disease characterised by long immunological, clinical latencies and chronic progressive inflammatory pathology. Multiple genomic regions associated with infection were reported indicating genetic complexity. In this study, a combined genome-wide approach (GWAS, Fisher's test and FST) using a high-density SNP array has been performed, comparing VMV-infected (n=78) and non-infected (n=66) individuals of the Valle del Belice sheep breed. The serological tests showed minimal variations in seroprevalence across the eight investigated herds, averaging at 26%. The comparison among the results revealed two association signals: on OAR03 close to the GRIN2B gene and on OAR05 close to the TMEM232 gene. The GRIN2B gene plays a role in pain response, synaptic transmission, and receptor clustering, while TMEM23 is involved in the development of immune-related disorders. The results highlighted new aspects of the genetic complexity related to the resistance/susceptibility to VMV in sheep. Additional analysis would be necessary to validate our findings.

ARDI2: Towards the consolidation of joint work in research, development and innovation in sheep

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Historically, the exploitation of the indigenous and similar dairy sheep breeds Latxa and Manech has been separated over time, and has led to the implementation of independent breeding schemes. The need to move towards the unification and the common management of the breeding programs was addressed in a previous POCTEFA project called ARDI, where work was carried out around the weaknesses, exchange of information and comparison of methodologies. As a result, the European Aggrupation of Economic Interest ARTALDEAN was created between Spanish and French farmers with the aim of support and strengthen the common management of the genetic programs. The aim of the new project ARDI2 is make of ARTALDEAN a reference organization for the implementation of common genetic actions on both populations. New models of international genomic evaluation will be defined, methodologies of diversity analysis to evaluate how genetic exchanges have been influenced the genetic variability will be evaluated, and traits related to sustainability will be defined. In addition, the incorporation of new technologies to the data recording will be studied, and different production systems will be considered. It is expected that results of ARDI2 support the strengthening of the dairy sheep sector in the Pyrenees and promote the competitiveness of Latxa and Manech breeds.

GEREMES: a project to improve the genomic insight on the resilience background in Mediterranean sheep breeds

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The ability of breeds to cope with not optimal environmental conditions is important for the livestock sector, especially in the regions of the World already affected by the climate changes. This is the case of the Mediterranean area, where the agriculture is already limited by climate change. Sheep breeds represent an ideal model to analyse the environmental adaptation for livestock species. Thus, it would be important to identify some indicators of resilience and climate adaptation. Based on these considerations, the project entitled “Genomic study of the resilience background in Mediterranean sheep breeds” with GEREMES as acronym aims to identify genomic regions associated with environmental adaptation. The main objectives are: i) to study the genome of sheep raised in different environmental conditions, to highlight genomic regions associated with these features; ii) to identify resilience indicators through machine learning and multivariate approaches; iii) to compare the genotypic data of local sheep raised in South Italy with cosmopolitan and with breeds from the North of Europe. We will use a multi-disciplinary approach combining quantitative genetics, population genomics, phenotypic and environmental data. The impact of this proposal encompasses important aspects hinged on many current topics that require urgent attention such as the use of integrated and sustainable natural resources, provide innovative knowledge to address the issues related to climate change and the maintenance of local biodiversity, with consequent conservation of traditional multifunctional landscapes.

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Poster 16

Insight into the genetic composition of the South African Bonsmara breeds using whole-genome sequencing data

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The Bonsmara is numerically the largest beef breed in South Africa and are present in stud breeding, commercial cow-calf operations, and feedlots. The breed makes use of an upgrading system, implying unknown base composition. South African cattle breeds such as the Bonsmara present an important source of genetic variation as it's an adapted breed with high meat quality. The objectives of the study were to assess the genomic diversity and population structure of the Bonsmara, Afrikaner and Shorthorn cattle that were base breeds in the development. Whole genome sequence data of the Afrikaner (n = 42), Bonsmara (n = 43) and Shorthorn (n = 20) were run through GATK's Best Practices pipeline and a jointly called vcf file was produced. Following standard quality control with the PLINK v2.0 software, a total of 367 764 autosomal single nucleotide polymorphisms were retained for further downstream analysis. The average heterozygosity values for the animals were highest in the BON (0.379), followed by the AFR (0.329) and the SHH (0.305). Moreover, the inbreeding coefficients for the animals were 0.154, 0.026 and 0.174 for the AFR, BON and SHH, respectively. Genomic structure analysis revealed that the BON have an admixed genome between the AFR and SHH, confirming the development of the breed. Hereford cattle were also used as a base breed, but no sequence data was available for this study. AFR cattle has been using BON in selection programmes and genetic infusion was clearly visible in some of the AFR cattle. A comprehensive genomic characterization of the Bonsmara and its base breeds using WGS data is necessary to detect genome level diversity and ancestry, and to assess breed composition over time.

Genomic evaluation for persistence of lactation in Girolando cattle

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Genomic evaluations were carried out using 784,093 test day milk yield records from first lactation Girolando cows spanning from 1998 until 2021. The aim was to identify a suitable model for future lactation persistence genomic evaluations. Genomic breeding values were predicted using random regression models with different Legendre polynomial functions through the best genomic unbiased linear predictor in a single step (ssGBLUP) approach. Following this, ten different measures of lactation persistence were evaluated. The model employing fourth-order Legendre polynomials, along with fixed effects of contemporary groups and heterozygosity, was identified as the best fit to the data based on the goodness-of-fit criteria. Heritability estimates for persistence ranged from 0.08 to 0.69. Genetic correlations between persistence and 305-day milk yield records varied from low to high magnitude (-0.04 to 0.96). The average genomic estimated breeding values (GEBV) of sires for the different persistence measures ranged from -41.46 to 249.70 kg. In conclusion, the measure of persistence represented by the sum of GEBVs considering the period from 55 to 270 days of lactation, obtained as GEBVs deviations at 50 days of lactation, should be preferred in genomic genetic evaluations for this trait in the Girolando breed.

Session 31

Poster 18

Genetic Characterization and Pathway Analysis of Carcass Weights in a Jeju Native Black×Hanwoo Crossbred Population

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Jeju Island, which boasts a unique environment due to its isolation, has historically seen the Jeju Native Black (JNB) cattle playing an indispensable role in the island's agrarian society. Despite facing endangerment during Japanese occupation and agricultural modernization, the JNB population surged from 31 in 1994 to 1,611 in 2016; however, the current population stands at 1,087. This study investigates genetic traits crucial for the preservation of JNB, utilizing a genome-wide association study and examining biological pathways in a Jeju-huekhanwoo (JHH, Jeju Native Black×Hanwoo) Cattle. The analysis, based on 39,055 SNP markers from 256 JHH cattle, identifies six quantitative trait loci (QTLs) associated with carcass weights on Bos taurus chromosomes 3, 5, 6, 10, and 13. The genes EIF2B3 and HECTD3 on BTA3, SOX5 on BTA5, ENSBTAG00000064813 on BTA6, ENSBTAG00000064392 on BTA10, and KLF6 and PHACTR3 on BTA13 were identified as positional candidate genes for these QTLs. Moreover, the identified positional candidate genes linked to carcass weights were enriched in pathways related to lysine degradation, axon guidance, tryptophan metabolism, glycerolipid metabolism, fatty acid metabolism. Future verification studies of these identified SNPs and positional candidate genes in other cattle populations hold the potential to provide valuable insights into the genetic architecture of carcass weights within Jeju Native Black cattle and Hanwoo-related populations.

Selection signature analyses revealed multiple genomic regions associated with milk production in Girgentana breed

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The Girgentana goat is an ancient Sicilian breed with distinctive morphological, adaptive and production traits. In this study, the integrated haplotype score (iHS) was used to detect within-population selection signatures in a sample composed of 205 Girgentana animals. Then, the haplotypes from these individuals were compared with those from other Italian goat breeds (5 from Southern and 8 from Northern Italy) to detect genomic regions under divergent selection, using Rsb approach. After editing, 536 animals and 48,744 SNPs were available. Multidimensional scaling and the individuals' Neighbor joining tree revealed a clear separation of three major clades coinciding with Girgentana, the Northern and Southern Italian breeds. A total of 14 genomic regions were detected: three within the Girgentana (iHS) and eleven from the comparisons Girgentana vs Northern and Girgentana vs Southern breeds (Rsb). The largest overlap between the two approaches was on chromosomes 1 (110 – 112 Mb) and 5 (38 – 40 Mb). Among the several selection sweeps, worth noting the region on chromosome 6 mapping to the casein genes (CSN2, CSN1S1, CSN3) known to be involved in milk quality. Other selection signatures mapped genes associated with body size, reproduction, and immune resistance. The results presented here provide a foundation for detecting mutations that underlie genetic variation of economic important traits for breed.

Genomic analysis of longevity with productive and reproductive traits in Girolando cattle

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The objective of this study was to estimate (co)variance components, heritability and genetic associations between different measures of longevity, productive and fertility traits in Girolando cattle using genomic selection. Phenotypic, pedigree and genotype records were obtained from the Brazilian Association of Girolando Breeders. A sample of 24,993 animals was genotyped with chips of different densities. Longevity measures related to the productive life of the cow, as well as productive and fertility traits were evaluated. The analyses were carried out using the AIREMLF90 software. Heritability estimates for measures of longevity, productive and reproductive traits ranged from 0.06 to 0.31, 0.06 to 0.13 and 0.03 to 0.03, respectively. Genetic correlations between longevity measures and productive traits ranged from 0.02 to 0.31, while genetic correlations between longevity measures and reproductive traits ranged from 0.00 to 0.13. Productive traits showed a greater genetic association with longevity measures compared to reproductive traits, suggesting that genetic improvements obtained in milk production can lead to longer retention of cows in the herds. Longevity measurements were found to be significantly influenced by the environment, indicating a low potential for response to selection. Therefore, they should be combined with other traits in an index to achieve greater genetic gains.

Calvana breeding scheme using genomic information and optimal contribution selection

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In conservation programs for endangered breeds, recording genealogies is essential for managing population inbreeding. Recently, the management of inbreeding, and consequently the management of matings, can be carried out through genomic information. These two different approaches have different limitations and opportunities. The present study aims to evaluate the differences in genetic diversity resulting from using either pedigree or genomic data to manage matings in the Calvana cattle breed, considered to be at high risk of extinction. Genealogical information on the Calvana breed was obtained from ANACLI. Moreover, 218 Calvana animals (35 males, 183 females) were genotyped with GeneSeek GGP-LDv4 33k; then, the genomic relationship matrix was created using the VanRaden method (2008). Breeding plans were developed using EVA software v.3.0 using the pedigree relationship matrix (P) and the genomic relationship matrix (G). Four scenarios were developed: two for the pedigree approach and two for the genomic one. All females were used in each scenario, while the maximum number of matings for each bull was set to 6 (Ped6, Gen6) and 20 (Ped20, Gen20). All candidate bulls (35) were selected as breeders, and no significant differences were found in the average inbreeding of the progeny between the four scenarios (min 0.0046 and max 0.0052). Using pedigree or genomic relationship matrix leads to differences in the number of matings per sire: it became higher when increasing at 20 the maximum number of matings for each bull. The different approaches resulted in quite different specific combinations between males and females: 97 out of the 183 matings differed between Ped6 and Gen6, and 93 out of the 183 matings differed between Ped20 and Gen20. To improve breeding plans, it is desirable to genotype a more significant part of the population to produce more accurate inbreeding estimates obtained by an improved genomic relationship matrix. This study was carried out within the Agritech National Research Center (Spoke 7) and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4–D.D. 1032 17/06/2022,CN00000022)

Session 31

Poster 22

Genomic characterization of the Montanina cattle

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Here we provide the first genomic investigation on Montanina, an ancient local dual-purpose Sicilian (Italy) cattle. Montanina is typically reared using extensive systems and adapted to the harsh mountainous environment. However, anecdotal knowledge of crossbreeding with cosmopolitan breeds to improve profitability exists. After quality checks, 925 animals from 37 Italian and French cattle breeds, including 35 Montanina individuals from three farms, were genotyped using medium-density SNPchips. The inbreeding levels were determined through Runs of homozygosity-based statistics. Population structure was analyzed through multidimensional scaling (MDS), Admixture, and Reynolds and identity-by-state (IBS) distances. We recorded low levels of inbreeding comparable among the three Montanina populations. Both IBS and MDS identified an intermediate position of Montanina between beef-selected breeds and other Sicilian breeds. By investigating the ancestry components in higher detail, we identified Limousin as the main source of introgression in Montanina. Further, we recorded lower levels of introgression in one of the three sampled farms. Our results reflect the selection history of the breed, including the acknowledged crossbreeding with Limousin bulls, aimed at increasing meat yield. These findings provide instrumental insights into the Montanina's genetic structure, diversity, and history, to guide breeding efforts balancing productivity and breed originality preservation. Work supported by CoRFilCarni through the BIOSAVE project.

Evaluation of genetic diversity in beef cattle breeds of Centre-Italy

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The value of the beef cattle breeds of Central Italy lies in their distinctive genetic heritage, as they are part of the Podolian breeds, characterized by unique white-grey coat and high rusticity, with origins that are subjected to debate. Additionally, their intrinsic connection with the culture and economy of their rearing area further emphasizes their significance. The genetic diversity of these breeds has not totally discovered yet, and the present work aims to analyse some new insights on this issue, thanks to the availability of medium density (33k) SNP genomic information on Chianina (CH, n=4,800), Marchigiana (MRC, n=4,700), Romagnola (RO, n=2,300), Maremmana (MRM, n=1,100), and Podolica (PO, n=2,900). The hierarchical structure of the populations and genetic distances were examined using the Admixture software. Selection signatures were then investigated by identifying runs of homozygosity (ROHs) for each breed (DetectRuns, R Package). ROHs were then used to calculate genomic inbreeding and identify genes within or near to the ROH regions. Results revealed that each breed represent a unique genetic pool. However, rustic breeds, MRM and PO, share a closely related genetic background, while MRC includes part of the genetic variability observed in CH and RO. ROHs were identified across various chromosomes, including 5, 7, 13, 18, and 21, encompassing genes associated with age at the first parity, docility, sexual maturation, regulation of RNA expression, and immunity defense against pathogens factors crucial for individual fitness. These findings shed the light on the genetic factors underlying the high rusticity of these breeds, a main factor of their evolutionary success. This work has been realized thanks to the iBeef2 project – PSRN Sottomisura 10.2 2014-2020.

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Poster 24

Genomic Landscape of a Newly Unveiled Meleagris gallopavo Population in South Italy: A Leap Toward Understanding Turkey Biodiversity

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Italy's indigenous turkey populations origins tracing back to the early specimens from the Spanish colonies in Central America. Notably, the turkeys' introduction to Italy in 1520 and their rapid proliferation, particularly in the south, underscore their integration into Italy's agricultural and culinary heritage. Other bone remains suggest that it was also used for food starting from 1600, so much so that it is found described in culinary recipes of the time. We conducted a genome-wide analysis of 5 small turkeys' nucleus (a total of 72 samples) reared in marginal area of Lecce (Puglia) and Potenza (Basilicata). DNA will be extracted and sequenced (12X). Data from other 11 local Italian population have been utilized: comparative analysis of 11 local Italian turkey populations: Brianzolo, Bronzato, Colli Euganei, Ermellinato di Rovigo, Ibrido Commerciale, Narragansett, Nero Italiano, Parmense and Romagnolo. Our findings, based on the genetic diversity indices—expected heterozygosity, observed heterozygosity, minor allele frequency, and inbreeding coefficient, reveal distinct genetic clusters, suggesting unique population structures and historical lineage continuity. The Fis values (0.275 ± 0.0245) suggest a high level of inbreeding or as the results of the PCA and Admixture would suggest, a strong isolation effect between the herds. By understanding the genetic makeup of these indigenous breeds, we pave the way for informed conservation strategies, ensuring the preservation of Italy's rich agricultural biodiversity for future generations. This study is part of the activities financed by the BIO.ZOO.CARE project: “PSR Puglia 2014-2022. Misura 10 – Pagamenti agro-climatico-ambientali. Sottomisura 10.2 – Operazione 10.2.1”

Next-Generation sequencing errors lessens the estimation of recent inbreeding

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Recent inbreeding is detected by identifying long stretches of homozygosity in Runs of Homozygosity (ROH). Next Generation Sequencing (NGS) improves the accuracy in the estimation of the length of ROH but it is prone to errors. Some 1.2 million of sequence errors are expected in a genome of 1.2 Gb (*Gallus gallus*) at a sequencing error rate of 0.1%. A large number of those errors will fall within ROHs, consisting of changes from homozygous to heterozygous bases and a breakdown of larger ROHs into smaller fragments. DNA from 20 hens of the autochthonous breed of laying hens Castellana Negra from the Conservation program at INIA-CSIC El Encin (Spain) were sequenced. After assembling with reference genome GRCg6a, inbreeding coefficients (total ROHs over total number of sequences) ranged from 0.31 to 0.47. There were 328,741 ROH fragments with 67 ROHs larger than 1Mb, and 96.4% of all ROH were 100kb or less. This abundance of small ROH fragments suggests that sequencing errors may reduce the estimation of recent inbreeding (large ROH fragments). We modeled sequencing errors as a Poisson process in which an error has a probability of happening at a constant rate across the genome. In this model, inter-arrival times are the length of the ROHs, which follows an exponential distribution. Fitting an exponential distribution to the length of ROHs of the 20 hens gave a rate of 0.005 (9.4e-06). This analysis only includes ROHs with polymorphic SNPs.

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Poster 26

Bayesian Genome-wide Association Study of udder-type traits in Alpine Grey Cattle

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The morphological conformation of udder and teat structure, given its high medium heritability, can serve as a proxy trait to select cows more resistant to mastitis. However, the biological implications regarding the connection between udder health, mastitis infection, and milk production have been thoroughly investigated. This gap of knowledge is even more pronounced in less specialized breeds. The study aimed to detect associations between genomic regions and udder-type traits in the local Alpine Grey cattle. After imputation, we used a 110K single nucleotide polymorphism (SNP) markers panel to profile the genotype of 1,194 individuals to investigate udder depth, fore udder strength, fore udder length, suspensory ligament, teat position rear view, and teat length. A single marker regression model corrected by population structure was used to test the associations of each SNP. De-regressed EBVs of the target phenotypes were calculated on 36,309 animals using a mixed linear model. Bayesian GWAS was performed by using the de-regressed EBVs of genotyped individuals. Utilizing the UCSC Genome Browser, we inspected the genomic position of selected markers using a Bayes factor greater than 100 as a threshold, assessing marker's relationship to genes or other functional DNA elements. The genomic regions associated with udder-type traits were spread on BTA1, BTA10, BTA12, BTA16, BTA17, and BTA18. The results led to the detection of well-known and novel genes for milk production (e.g., C4BPB, PEX14), mastitis resistance (e.g., PIAS1, TLR2), and mammary epithelial development (e.g., TBX3, SPRY1). The biological pathways of these genes have been also investigated. Further studies should also consider the relationships with other production and fitness traits. Acknowledgements: Funded within PSRN – Dual Breeding 2 (J51J1800000005), and within the “Agritech” – Next-Generation EU PNRR – Missione 4 Comp. 2, Invest. 1.4 – D.D. 1032 17/06/2022, CN00000022.

Italian deli meats quality perspective with focus on best practices and commitments towards a new business model

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ASSICA, the Industrial Association for Meat and Deli Meats, is the national trade association representing pork slaughter and processing companies. Italian Deli Meats are an international success: Data for 2023 show record exports both in terms of quantity and value. These figures underline the economic and qualitative value of this agri-food sector. With no less than 43 types of PDO and PGI-certified Deli Meats, the industry is also Europe's record holder for protected products. The quality of pig meat has evolved over time under the influence of several genetic, environmental and food factors. Thanks to selection and research in the field of animal nutrition it has been possible to further improve the nutritional profile of pig meat making it more and more adequate to meet the needs of modern lifestyles. As a consequence, it is true that Italian deli meats available on the market are no longer as they used to be, but they are better: Less fats and more proteins; Less Salt; More Vitamins and Mineral salts. Furthermore, ASSICA is now setting new environmental, economic and social objectives to develop the sector while respecting its distinctive traditions. ASSICA with IVSI (Institute for the promotion of the Italian Deli meats) has already presented the «2022 sustainability programme» – and is now working on the 2024 update – of the Italian deli meats sector for companies to improve their contribution to the achievement of the Sustainable Development Goals. This publication is the first collection of best practices, corporate and association commitments and actions towards sustainable development, and a further and tangible step in the path that ASSICA is building together with Member Companies to promote an entrepreneurial approach based on sustainability, understood in all its meanings (environmental, economic and social). Indeed, what is underway is a process of acquiring awareness and consequent actions, based on a cultural step-change where sustainability is a business opportunity and not a burden, as a requirement to remain competitive on the market. The evolution of this approach means considering sustainability as one of the central factors, values, of present and future corporate strategies.

Understanding the development of intramuscular fat in cattle – a mini review

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Intramuscular fat (IMF), also known as marbling, results from adipocyte proliferation and expansion within connective tissue and vascular seams of muscle. The stored triacylglycerol has a positive influence, even at moderate levels, on the sensory properties of beef. The expression is heavily influenced by the extend of muscle growth given that IMF is the ratio of [fat]/[fat + muscle]. So, younger animals, more muscular breeds and entire males have lower IMF levels due to muscle development diluting fat accretion. In addition, muscle groups with more aerobic muscle fibres have generally increased level of IMF. A key enabler of IMF expression is genetic selection for this trait specifically, rather than for fat deposition at other sites such subcutaneous fat. There is consistent evidence that IMF adipocytes are different to those found at other depots with the cells being smaller, metabolically less active and typically showing a preference for glucose and lactate as a fat precursor rather than acetate. Consistent with this, is the role of feeding high energy diets (typically containing starch from cereal grains) to improve the expression of IMF. However, nutritional manipulation of IMF independently from other fat depots has proved more problematic with typically total carcass fatness also increasing along with IMF; although manipulating the Vitamin A axis has shown some promise. There is an increased expression of IMF in older cows at a given total body fatness and muscularity which may relate to fat turnover favouring IMF over many calving cycles. The higher IMF in cows is an opportunity to value-add selected cuts to meet consumer expectations. A French premium beef brand was launched to better value IMF.

Transforming chemical intramuscular fat% into MSA and AUS-MEAT marbling scores and potential application in the Australian beef industry

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Meat Standards Australia (MSA) and AUS-MEAT marbling score are key determinants of carcass value in the Australian beef industry and are well recognised traits in national and international markets. However, with the emergence of objective measurement technologies there are opportunities to grade beef carcasses using objective traits such as chemical IMF%. The challenge is that this abrupt change to MSA model inputs and AUS-MEAT grading practices would cause significant disruption to the beef industry. A proposed solution is to develop an industry approved equation to convert IMF% into MSA marbling and AUS-MEAT marbling scores. These converted marbling values would then seamlessly integrate into existing grading and marketing systems, whilst being underpinned by an objective, gold standard trait. Carcasses ($n = 5513$) from industry experiments across 7 years (2017 – 2023) were graded for MSA marbling and AUS-MEAT marbling and sampled for chemical intramuscular fat (IMF%). Using this data, equations were derived to convert IMF% to MSA marbling (IMF%-MSAMB) and AUS-MEAT marbling (IMF%-AUMB). When validated the IMF-MSAMB model described 91% of the variation in MSA marbling (Residual Standard Error (RSEV) = 57.9), with a slope of 0.90 and negligible bias of -0.54. Similarly, IMF-AUMB described 88% of the variation in grader AUS-MEAT marbling scores (RSEV = 0.68) with a slope very close to 1 (0.94) and little bias (0.06). Industry-wide implementation of this equation is currently being considering in Australia. This would enable grading technologies to be calibrated and validated against chemical IMF% whilst minimising industry disruption.

Session 32

Theatre 4

Prediction of consumer palatability in beef using visual marbling scores and chemical intramuscular fat percentage

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With development of objective technologies that can predict chemical intramuscular fat percentage (IMF%), there is a need to understand the relationships between existing marbling traits, IMF% and eating quality. This presentation will report on an analysis of historical carcass data ($n = 9641$ observations) from the Meat Standards Australia (MSA) industry research dataset and included MSA grading data, chemical IMF% data and weighted composite eating quality scores (MQ4). Several analyses were performed to assess the prediction of eating quality by MSA marbling, *M. longissimus thoracis et lumborum* (striploin) IMF% and cut specific IMF%. Results demonstrated that there was similar precision between chemical IMF% ($R_2 = 0.32$, RSE = 11.8) and MSA marbling ($R_2 = 0.28$, RSE = 11.9) in the prediction of grilled 14 day aged striploin MQ4. These results support the development of objective technologies that predict chemical IMF% in parallel with MSA marbling for carcass grading and the prediction of eating quality.

Flavour drives consumer eating quality acceptability as intramuscular fat increases in Australian lamb

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Untrained consumer-determined overall liking, flavour, tenderness and juiciness are highly correlated due to consumers' inability to discriminate between these traits. Overall liking is an indicator of a consumer's overall product acceptability; hence, overall liking is considered to reflect flavour, tenderness and juiciness attributes. Increasing intramuscular fat (IMF) improves all eating quality traits, principally juiciness and flavour. Carcasses with a higher loin IMF% are juicier and more flavoursome; therefore, these traits could be expected to contribute more to the overall acceptability. The knuckle, loin, outside, rump, topside, leg, rack and shoulder were collected 24 hours post-slaughter from 3119 lambs. Each cut was assessed for overall liking, flavour, tenderness and juiciness on 100-point scale lines (100=best) by untrained consumers. Chemical IMF% was determined for the loin. Consumer responses (n=108410) were arbitrarily split based on IMF at 2.0-4.0, 4.1-6.0, 6.1-8.0 and <8.0%. Within each IMF category, data was divided into five subgroups balanced for IMF%. Within each subgroup, a linear model was used to predict overall liking, with flavour, tenderness, and juiciness simultaneously included as independent variables. Relative weight analysis then determined the contribution of each variable to the prediction of overall liking. The mean of the relative weights for each of the five subgroups was calculated and compared between IMF categories via a one-way analysis of variance. Flavour was the highest contributor to overall liking within each IMF category, improving ($P<0.05$) from 43.4 to 47.3% across the 10.8% IMF range. This was associated with a decline ($P<0.05$) in the contribution of tenderness (29.1-26.5%). The contribution of juiciness to overall liking was not affected ($P>0.05$) by IMF. The increasing importance of flavour between IMF categories has occurred over a relatively small IMF% range of as little as 4%. This implies that the contribution of flavour to eating quality may become more important as future genetic selection broadens the range of IMF% in lamb.

Session 32

Theatre 6

Effects of live yeast and selenium supplementation on rumen parameters in long-transported Charolais bulls
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Long-distance transport and receiving at fattening units are stressful for beef cattle. This study tested live yeast and selenium supplementation (Se) on 80 Charolais bulls transported from France to Italy. The bulls were randomly divided into Yeast and Control groups (40 animals each). Yeast group bulls received live *S. cerevisiae* CNCM I-1077 and Se-enriched yeast nutritional boluses before leaving France, and yeast in their diet in Italy. Rumen parameters (rumination, temperature, and pH) were monitored every 10 min using sensors in 60 bulls evenly distributed between the two groups. The reticulo-rumen pH was normalized, and daily mean, amplitude, standard deviation, and incidence of sub-acute ruminal acidosis (SARA) were summarized for the first and fourth weeks. Rumen indicators at the receiving to the fattening unit were analysed with a linear mixed model, considering supplementation group, age, day, and truck as fixed effects, and bull nested within pen as random effect. Yeast stabilised the rumen environment by limiting daily pH amplitude, standard deviation, and the inter-animal variability around transportation. The incidence of SARA was reduced in the Yeast group during the first week. Ruminal temperature was not impacted by the supplementation, but live yeast increased rumination time after the first month. Maintaining stable ruminal conditions is crucial for the well-being of beef cattle during diet transitions at fattening units. Supplementing cattle with live yeast before transportation could help them cope with ruminal stress upon arrival.

An evaluation of muscle cut, maturation and meat quality of beef using Rapid Evaporative Ionisation Mass Spectrometry (REIMS) analysis

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Rapid Evaporative Ionisation Mass Spectrometry (REIMS) is a rapid ambient analytical technique. Applications of REIMS include determination of biological tissue, food fraud and more recently meat quality parameters of beef. Determination of beef muscles, maturation period as well as the sensory quality using consumer taste panels and the Meat Standards Australia (MSA) grading system is currently unknown. Therefore, the aim of this study was to use REIMS and advanced chemometric modelling to uniquely connect REIMS data from four raw beef muscles, maturation, consumer sensory scores and MSA grading. Beef samples (N=149) from four muscles, were collected from 31 carcasses, processed with two hang methods, after 7- and 21-day maturation post-slaughter. Carcasses were MSA graded, and consumer taste panels were conducted at 18 different locations. Beef samples were burned using a monopolar electrosurgical "iKnife" attached to the REIMS system (Waters REIMS source (Waters Corporation, Wilmslow, UK); a Xevo G2-XS QToF Mass spectrometer (Waters Corporation, Wilmslow, UK). Data were performed in sensitivity mode with continuum data acquisition in negative ionisation mode REIMS data was extracted using the Abstract Model Builder software and analysed using Simca software. REIMS analysis successfully identified four beef muscles, in addition to 7- and 21-day maturation. Consumer taste panel results and MSA grading scores were also positively discriminative using REIMS. This study has demonstrated that REIMS can detect and categorise unique chemical fingerprints related to muscle, maturation, and sensory quality of beef.

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Theatre 8

REIMS (Rapid Evaporative Ionisation Mass Spectrometry) prediction of beef sensory quality and its relationships to fat level

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To characterize beef eating quality and its relationship to marbling and lipids, 31 young grass-fed crossbred Angus x Salers were used in this study. Three muscles (longissimus dorsi, triceps brachii, and internal abdominal obliques) were used for sensory evaluation and REIMS analysis. Results indicated that grass-fed crossbred Angus x Salers produced lean meat of "better than average" palatability. Although no significant correlation was observed between beef sensory quality and marbling/lipid levels, however, lipid molecules were the primary component detected by REIMS, REIMS showed great potential to classify beef sensory quality and grades (i.e., higher flavor liking vs. lower flavor liking; satisfactory vs. unsatisfactory) with accuracies up to 99%. In addition, REIMS has indeed potential for predicting visual-measured marbling and gas chromatography measured lipid and/or FA levels, with varying efficacy due to distinct REIMS fingerprinting. Overall, REIMS classification results may be more robust with larger sample sizes and wider variances.

A cuts-based Meat Standards Australia grading system for sheepmeat

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A new cuts-based Meat Standards Australia (MSA) eating quality grading system is being developed. This system requires objective measurements of carcass traits acquired at processing that describe eating quality variation. Previous studies have identified these carcass traits to be intramuscular fat % (IMF%), lean meat yield (LMY), and hot carcass weight (HCWT). To develop the model, data was collected from 3106 lambs of which 11040 cuts were sampled. Consumer tasting sessions (n=312) were carried out with 18720 untrained consumers who scored samples for tenderness, juiciness, flavour liking and overall liking on a scale from 0 (worst) to 100 (best). Loin, topside, outside, rump and knuckle cuts were grilled, whereas the rack, leg, shoulder, and knuckle cuts were roasted. A discriminant analysis of the 4 eating quality traits defined their combined weightings within a single meat quality score. A linear model was then fitted to predict this score, using IMF%, LMY, and HCWT as covariates and cut as a fixed effect. Results indicated that eating quality was predicted with an accuracy of 75% (grill) and 72% (roast), though great individual consumer variation exists. The model will allow allocation of cuts according to eating quality grades.

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Theatre 10

Exploring the meat quality of Lidia heifer meat: preliminary results

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Extensive systems with local breeds play a crucial role in preserving resources and ecosystems, as well as producing meat with unique characteristics. These attributes are in line with the current demands of European meat consumer. However, these models face challenges such as lack of profitability or substitution by more productive livestock alternatives. It is in this context that the study of Lidia breed meat arise. This is an autochthonous Spanish breed developed in an extensive system linked to the “dehesa” ecosystem. The meat of these animals has always been considered a secondary product, which is why there is an important scientific gap. In this work we intend to carry out a first analysis of the instrumental quality of the meat of Lidia breed females. A total of 200 heifers (24-48 months old) of the Lidia breed from 10 farms (20 animals per farm) were used. The animals followed a standard production cycle. From each animal the m. Longissimus thoracis, corresponding to the 12th-13th thoracic vertebrae, was obtained and the meat was vacuum aged under refrigerated conditions for 21 days. The variables analysed were pH, colour in CIELab* space, water holding capacity (thawing, drip, pressure and cooking losses) and texture (maximum shear force, WBSF test). Descriptive statistical analysis was applied using XLStat software. Lidia heifers showed an optimal pH, dark, red and moderately yellow meat colour. Acceptable water holding capacity and acceptable to intermediate tenderness were observed. The results found show that meat from Lidia breed heifers could meet industry and consumer demands.

Characterisation of Meat Quality Attributes in Different Cattle Breeds: Implications for Labelling and Consumer Choice

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Consumer preferences for beef are increasingly driven by the desire for high hygienic, nutritional, and organoleptic quality. This paper investigates the impact of cattle breed on key quality attributes—colour, marbling, and tenderness—central to consumer choice. Six different bovine breeds were taken into consideration: German Red Pied (n = 63), Piemontese (n=117), Chianina (n=63), Angus (n=63), Friesian (n=18) and a Polish cross breed (n=63). The cut of meat analysed was the entrecôte, or boneless rib, obtained from the muscles located between the fifth and eighth rib of the loin. Colorimetric assessments, marbling evaluations, fatty acid profiling, and tenderness measurements, were conducted on meat cuts from each breed. Results revealed significant variations in colour, marbling, and tenderness among breeds. Statistical analysis of the data was carried out using StatView 5.0.1 software (SAS Institute, Cary, NC, USA) for the analysis of variance (ANOVA) with Fisher PLSD (protected least significant differences). Chianina, Friesian, and the Polish crossbreed exhibited distinct colour characteristics, with Chianina displaying notably brighter meat. Angus emerged as the most marbled breed, while Chianina and Piemontese showed lower marbling. Total lipids content correlated with visible marbling. Fatty acid composition varied, with Angus having higher omega-3 percentages. Tenderness assessments identified Angus and Friesian as the most tender breeds. The study's findings contribute to a proposed grading scale for colour, marbling, and tenderness, offering potential labelling infographics to assist consumers in making informed choices based on individual preferences and needs. These insights underscore the importance of breed-specific information on labels to enhance consumer understanding and facilitate more informed purchasing decisions.

Session 32

Theatre 12

Evolution of animal and meat research over the past 50 years

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This work aims to study the evolution of research in animal and meat sciences from 1974 to 2023. The Web of Science database was used to count the number of scientific articles published per 5 year period in these fields. In practice, the following keywords were used: 1) cattle, pig, poultry or sheep for animal science; 2) beef, pork, poultry meat or sheep meat for meat science. Results were analyzed according to “Web of Science categories” or “country/region”. In animal science, “Veterinary sciences” and “Agriculture dairy animal science” categories have been dominant over time, with a relative decline in both cattle and poultry, unlike to the stable trends in sheep and pig. “Food science technology”, “Microbiology” and more recently (from 1999) “Environmental science” categories have gained importance in all species. In meat science, “Food science technology” is the leading category for pork and beef (about 40-60% and 20-30% respectively), “Agriculture dairy animal science” is the leading category for poultry meat and sheep meat (about 40%) and “Veterinary sciences” is the second one for sheep meat and occupies a lower position in other species. Over 50 years, the USA has led research across all areas, except sheep meat, ranking second behind Australia. China emerged in all fields after 2004, reaching top positions in 2019-2023 with the exception of beef. Similarly, research in Brazil developed earlier and has developed roundly since then. Canada is also a major actor except for sheep and sheep meat, while Germany and England have been players in all animal sciences. Overall, this work provides valuable insights into evolving trends and offers guidance for future research directions in animal and meat research.

Meat quality for once-calved heifers and heifers of Holstein-Friesian × Limousine crossbreds

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In Poland, approx. 200 thousand heifers are slaughtered annually. In view of the reduction in the population of calves which are suitable for fattening, a certain solution is to use the so-called once-calved heifers for the production of calves. Young growing cows, as compared with adult suckler cows, convert more feed energy to body weight gain, and therefore, for biological reasons, the technology of the use of once-calved heifers is very effective. The objective of the study was to compare the meat quality of heifers (n=16) and once-calved heifers (n=15) of Holstein-Friesian × Limousine crossbred slaughtered at the age of 18 and 28 months, respectively. The once-calved heifers at the age of 15 months, were bred with semen of Limousine bulls and following the calving they fed 100 days their offspring and then they were slaughtered. Meat of once-calved heifers contained more intramuscular fat, was darker ($P \leq 0.01$), and exhibited a higher water-holding capacity and lower cooking loss ($P \leq 0.05$) as compared with heifers' meat. In sensory evaluation meat of heifers was more tender ($P \leq 0.01$) and tastier as compared with once-calved heifers' meat ($P \leq 0.05$). Gestation and nursing had a negligent effect on the content of functional components in the meat of once-calved heifers, in comparison with the meat of heifers. In conclusion, due to the possibility for obtaining additional offspring and only a slight reducing in the sensory value of the meat quality, it is recommended to use heifers originating from commercial crossbreeding as the once-calved heifers. „Funded by the Minister of Science under „the Regional Initiative of Excellence Program”.

Session 32

Poster 14

Impact of immunocastration on meat quality and muscle transcriptome of Angus × Nellore cattle

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Intramuscular fat, also known as marbling fat, stands out as one of the most appreciated and desired attributes in meat, being positively correlated with its pricing. Twelve F1 Angus × Nellore steers (six non-castrated males and six immunocastrated males) were fed for 230 days in a feedlot and slaughtered at 16 months of age. The hypothesis of the study was that non-castrated male cattle would have meat quality equivalent to that of immunocastrated steers. The chemical composition of Longissimus thoracis was analyzed as well as the muscle transcriptome profile. Significant genes identified by gene coexpression analysis in the CeTF package of software R were considered transcription factors, and those with counts in all samples from one sex class and no counts in the other were considered exclusive. Immunocastrated steers showed greater deposition of intramuscular fat (non-castrated – 2.9% and immunocastrated – 6.5%; $P < 0.01$). There was no significant difference between groups for genes involved in lipogenesis, such as PPARG, FASN and ACACA ($FDR > 0.05$); however, the genes CPT1C (0.188) and SCP2 (0.003), involved in the β -oxidation of fatty acids, were identified as exclusive in non-castrated males. On the other hand, immunocastrated males showed higher expression of the genes PLIN1 (0.346) and FABP4 (0.565), responsible for the differentiation of adipocytes, identified as exclusive and transcription factors, respectively. Therefore the greater accumulation of marbling fat comes from the lower lipid oxidation and greater differentiation of fat cells.

Investigating tissue specific differences in the adipogenic cascade in bovine preadipocytes differentiation

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Adipose tissue development in farm animals can be modified by production factors, but it is not clear how to optimize marbling without increasing the total body fat mass. Regarding beef cattle, it is of interest a minimum intramuscular (IM) fat content to produce high quality meat while not increasing other adipose depots so feed efficiency does not decrease. But these are opposed goals as total body fat in the different depots is positively correlated. As adipocytes show tissue specific biological features, identifying depot specific traits leading to distinct adipose tissue development could favour those objectives. A bovine in vitro model of primary IM and subcutaneous (S) preadipocytes derived from 18 months old steers was used to investigate the expression of genes of interest in the adipogenic, antiadipogenic and lipogenic pathways, which were analysed in order to identify possible depot specific distinct features during differentiation. The induction of the genes of interest occurred in S adipocytes, in general, before in the time course than in the IM cells and the order of magnitude of the fold-change gene expression over undifferentiated cells was higher in S adipocytes. This was the case for PPARG and CEB-PA, master adipogenic regulators, while the key antiadipogenic factor WNT10B showed none or little variation; NR2F2, that could modulate both pathways, only was upregulated in S adipocytes. The downstream target genes FABP4 and LPL (exogenous fatty acids uptake) were upregulated in S and IM cells but ACACA and FASN (de novo fatty acid synthesis) were upregulated in S adipocytes. Those results suggest that NR2F2 and de novo fatty acid synthesis could be involved in the lower and retarded differentiation of IM adipocytes.

Session 32

Poster 16

On-farm preconditioning affects cattle stress, immune, and respiratory microbiota responses after transportation and feedlot placement

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Preconditioning programs are used to mitigate bovine respiratory disease (BRD) in feedlots, but limited information exists on how they affect immunity and respiratory microbiota. Objective: to test the effects of preconditioning on cattle stress response, immunity, and respiratory bacteria. Methods: farm calves were enrolled into preconditioning (PC, N=50; three immunizations, low-stress weaning) and non-preconditioning (Non-PC, N=50; single immunization, abrupt weaning) groups. Three weeks after weaning (or day of abrupt weaning for Non-PC), calves were transported to a feedlot and comingled with market calves. Nasopharyngeal (NP) swabs were collected before transportation, at feedlot arrival, 2, 6, 14, and 32 days after feedlot arrival, and before treatment if diagnosed with BRD. Blood was collected before and after feedlot placement. The 16S rRNA gene was sequenced from NP swabs and cortisol was quantified in blood. Pathogen-specific IgG or IgA in blood and swabs, respectively, were measured against Mannheimia haemolytica and Histophilus somni. Bacterial, immune, and stress variables were analysed in R studio. Results: cortisol did not change in PC calves ($P>0.05$), but increased after transportation in non-PC animals ($P<0.05$). Compared to non-PC calves, the PC group had increased *M. haemolytica*-specific IgG and IgA at feedlot arrival ($P<0.05$), while *H. somni* immunity did not differ. Morbidity due to BRD was 44% and 50% in PC and non-PC groups, respectively. In BRD cases, the dominant genera were Histophilus (26.4%), Mycoplasma (24.9%), Moraxella (7.2%), and Pasteurella (6.0%). The non-PC cattle had elevated Pasteurella, Histophilus, and Moraxella, compared to PC animals ($P<0.05$). Conclusions: preconditioning reduced stress response and the abundances of several BRD-associated genera in calves. Increased on-farm vaccination frequency improved host immunity at feedlot arrival, however the effect was variable for different pathogen targets. Overall, preconditioning enhanced respiratory health when calves experienced management stressors. However, improved vaccine efficacy may expand protection further.

Use of a commercial feed supplement based on diatom earth and yeast products on oxidative status and in vitro immune response in buffaloes during peripartum

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The aim of this study is to evaluate the effect of a commercial feed additive based on diatomaceous earth and hydrolyzed yeasts on health status, milk quality and immune response of buffalo cows during the transition period. Eighty healthy Italian Mediterranean buffaloes were included in the trial, subdivided in two groups: a group received the additive (n = 40) while the control group (n=40) received a placebo. Blood samples were collected from each buffalo at -60d (60 days from the expected calving), -30d (30 days before the expected calving), 0d (calving), +15d, +30d, and +60d (respectively 15, 30 and 60 days in milking). Biochemical, oxidative, antioxidant and enzymatic profiles were evaluated. Moreover, acute phase proteins, reactive proteins and Interleukine plasma levels were determined. Peripheral blood mononuclear cells (PBMC) and monocytes were isolated and viability, reactive oxygen species (ROS) and reactive nitrogen species (RNS) detection were performed on PBMC and monocytes. The additive administration did not affect both colostrum immunoglobulin and milk yield and composition during the early lactation period. It enhanced the total antioxidant capacity and enzyme activity, while no differences were observed in oxidation products. Despite higher enzyme levels in plasma, this didn't necessarily correspond to significantly increased enzymatic activity, but rather indicated a higher potential. The transition period in buffaloes differs notably from what reported in literature for cows, probably due to the absence of common postpartum production diseases in dairy cows and lower metabolic challenges linked to lower milk production in buffaloes.

Session 33

Theatre 2

Effects of a nutritional supplement in Italian Mediterranean Buffalo exposed to heat stress

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Heat stress (HS) has a negative impact on the production and quality of milk, on reproduction and on health of dairy ruminants including buffalo. To alleviate HS some strategies are available such as cooling systems. Otherwise, nutritional strategies can be a useful tool for improving animal wellbeing, milk yield and quality in buffaloes exposed to HS conditions. In this context this study aimed at investigating the effect of RuFO[®] supplement in lactating buffalo exposed to HS. Three groups of 19 buffaloes each were used: control group (C) received the basal total mixed ration (TMR), T1 and T2 received TMR supplemented with 100g/head/d and 200g/head/d of RuFO[®], respectively. The experiment started in early June and ended in early September for a total of 93 days. Individual milk yield (MY) was daily recorded, and milk samples were analyzed every two weeks. TMR and feces samples were taken and analyzed, nutrient apparent digestibility (DIG) was calculated, and animal behavior was constantly monitored by ear tag sensors. Data were processed by a nested design ANOVA in which "sampling" and "group" were set as fixed factor, and "buffalo" were set as random factor and nested into "group". T1 had higher MY with lower content of somatic cells and improved rheological parameters. DIG was improved in T1, and T1 spent more time in rumination and eating. In conclusion, the administration of 100g/head/d can be suggested as an adequate quantity of Rufo[®] that help animals to counteract HS and improved MY and quality.

Feasibility of visible/near-infrared spectroscopy to predict the chemical composition of feces and total-tract apparent nutrients digestibility in lactating buffaloes

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The potential of near-infrared spectroscopy (NIRS) to predict chemical composition of feces and nutrient total-tract apparent digestibility (TTD) in lactating buffaloes was evaluated. Individual fecal samples (n = 147) were collected in 5 farms during the BIG project (ANASB 2021). Animals were fed silage-based total mixed rations, which were sampled 2 days prior to fecal collection. Nutrients were estimated using both uNDF and AIA as internal markers (determined on diets and feces). Strong correlations (r) were observed between TTD of EE (0.96), hemicellulose (0.83), NDIN (0.91), ADIN (0.76), NB3 (0.93), NFC (0.86), and starch (0.91) estimated using uNDF and AIA as markers. To validate the NIRS prediction models, the dataset was divided into a calibration set (75%) and a validation set (25%) for external validation. Average accuracies, indicated by the coefficient of determination in external validation (R₂ExV), were achieved for fecal N, CP, and aNDF content, while excellent predictions were obtained for TTD of ADIN (0.90) estimated using uNDF as internal marker. Additionally, similar accuracy in predicting TTD was observed for EE calculated from uNDF (0.89) and for EE calculated from AIA (0.86). Results demonstrated the effectiveness of NIRS technology to predict many traits relate to quality composition and nutrient digestibility of fecal allowing the evaluation of nutrients status of animals. This research was funded by MIPAAF – DISR 07 – PSR 2014/2020 BIG” Prot. N. 0215513 11/05/2021.

Session 33

Theatre 4

Influence of an Automatic milking system on milk yield and quality in Italian Mediterranean buffaloes

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Scientific research on the use of automated milking system (AMS) in Italian Mediterranean buffaloes (IMB) is still limited. The aim of this study was to compare conventional milking and AMS, focusing on differences in milk yield (MY), quality and somatic cells count (SCC). The trial was carried out in a farm in Caserta province, equipped with both AMS and conventional milking. At calving, 100 IMB heifers were randomly assigned to two experimental groups: AMS Group (AG, n=50) and Conventional Group (CG, n=50). For each animal, daily MY was recorded, and a milk sample was collected monthly to determine fat percentage (FP), protein percentage (PP) and SCC. Statistical analysis was carried out using repeated measures ANOVA (SPSS Inc., Chicago, IL, USA). A higher (P<0.01) MY (1.05 kg/day) was recorded in AG compared to CG group, probably due to the higher number of milkings (2.34 vs 2 milkings/day for AG and CG respectively). Total production throughout lactation was 2,320.6 vs 2,036.4 kg in AG and CG, respectively (+12.2%). With regard to milk quality, no statistically significant differences were recorded in FP (8.24±0.7 vs 8.09±0.7) and PP (4.77±0.4 vs 4.80±0.3). On the contrary, AG had significantly lower SCC (P<0.01) than CG (49.92±15.5 vs 153.09±57.3, respectively). In conclusion, AMS in IMB exhibits a better efficiency compared to the conventional milking system.

Transcriptomic characterization of buffalo's extracellular vesicles from colostrum and milk: cargo comparison for potential applications

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Recently, much interest has been raised in the characterization of signaling molecules carried by extracellular vesicles (EVs), particularly enriched in milk (mEVs). EVs can cross biological barriers, resist acidification in the gastric environment, and exert modulation of the immune system, mainly through their microRNA (miRNA) content. This work aimed to characterize the small-RNA cargo of colostrum EVs (colosEVs) and mEVs of Italian Mediterranean buffalo through next generation sequencing (NGS). Seven subjects were selected for one sample collection of colostrum (first milking after parturition) and milk (day 50 of lactation). ColosEVs and mEVs were isolated through differential centrifugations, an EDTA treatment and ultracentrifugations, and total RNA was extracted and Illumina sequenced. The main difference was the amount of EV in the two samples, with colostrum showing 10 to 100-fold higher content than milk. For both types of samples, miRNA was the most abundant RNA class (95% for colosEVs and 96% for mEVs) and three lists were identified: a) shared most expressed, b) colosEV-specific, c) mEV-specific. For each list, targets were retrieved and a protein-protein interaction (PPI) network was built. The gene ontology (GO) enrichment analysis highlighted many terms related to the epigenetic, transcriptional and translational regulations, including protein phosphorylation for both colosEV and mEV miRNAs. Many biological processes related to immune modulation were found for both biological samples. Moreover, for sample-specific miRNA targets, ColosEV enriched terms were related to “cell differentiation” and “microvillus assembly”, while mEV showed biological processes for “cardiac and blood vessel development” and “mitochondria”. The whole message carried by the EVs from the two matrices is similar, but we observe a great difference in the amount of EVs, since colostrum shows a higher content than milk.

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Prediction of blood parameters of buffaloes from the milk mid-infrared spectra

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Blood metabolic profile testing allows for monitoring metabolic health and nutritional status. Still, conducting extensive blood analyses on a large scale is not feasible due to high costs, labour, and animals' stress. In this context, utilizing Fourier-transform mid-infrared (FT-MIR) spectra of milk to predict blood traits may present an effective opportunity. The present study aims to test the ability of milk FT-MIR to predict blood traits using milk spectra collected from the same buffalo. Blood and milk samples were collected from 310 buffaloes in different stage of lactation reared in 9 farms in the Southern Italy. The concentration of non-esterified fatty acids (NEFA), triglycerides, cholesterol, low-density lipoprotein (LDL), high-density lipoprotein (HDL), glucose, urea, total protein, albumin, globulins, creatinine, aspartate aminotransferase (AST), alkaline phosphatase (ALP), glutamic oxaloacetic transaminase (GOT), glutamic pyruvic transaminase (GPT), gamma-glutamyl transferase (GGT), creatine kinase (CK), lactate dehydrogenase (LDH) and total bilirubin (TBIL) were measured in blood through reference analysis. Milk samples were scanned with a MilkoScan and FT-MIR spectra were collected. Spectral data were divided into a calibration set (70%) and a validation set (30%) for external validation. In external validation, coefficients of determination ≥ 0.60 were achieved for urea, triglycerides, NEFA, LDL and TBIL concentrations. Although FT-MIR models cannot be considered accurate enough for punctual determination, the predictions of blood parameters can be considered for the screening of the herd and for genetic purposes at population level.

Cortisol concentrations in different matrices: predictive potential and relationship with productive level, lactation stage and parity in dairy buffaloes

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Cortisol is a biomarker related to hypothalamic-pituitary adrenal (HPA) axis activity. Plasma and milk cortisol concentrations provide short term evaluations about the HPA axis activity while hair can be used for the retrospective assessment of cortisol levels. This research aimed at studying the relationship among cortisol in blood, milk, whey and hair with parity, days in milk (DIM) and productive classes and to study their predictive potential. Multiparous (n=30) and primiparous (n=38) buffaloes were assigned to 4 productive classes, according to equivalent corrected milk (ECM) (ECM < 15.85; 15.86 < ECM < 17.57; 17.58 < ECM < 19.74; ECM > 19.75), and 3 classes of DIM (<90 DIM; 91<DIM<150; >150 DIM). Cortisol levels were measured by RIA method. Parity did not show an effect on cortisol concentrations. The catabolic stage of lactation (up to 90 DIM) was characterized by higher cortisol concentrations in milk compared to the second anabolic (beyond 150 DIM). Plasma cortisol concentrations were higher at the catabolic and the first anabolic stage (91 to 150 DIM) compared to the second anabolic (P=0.022 and P=0.009 respectively). Buffaloes beyond 150 DIM differed from those below 90 DIM (p<0.001) and between 91 and 150 DIM (p < 0.05) in hair. Hair cortisol concentrations were negatively correlated to mature equivalent milk yield (p<0.05). Finally, the whey showed a potential to predict the concentrations of cortisol in whole extracted milk (R²=0.31).

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Characterization of buffalo faecal microbiome depending on ration concentrate level

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The microbiome communities of the gastrointestinal tract (MCTGI) of ruminants plays an essential role in animal health and productivity. It is well known that diet has the greatest influence on MCTGI, with the potential to alter the microbial diversity. The aim of this study was to evaluate the effect of different concentrate level rations on the microbiota composition of buffalo faeces. Faeces samples (FS) were collected from 10 dairy buffalo farms in Lazio region (Italy) once a month, for 6 months. FS were collected and DNA was extracted using Faecal DNA MiniPrep kit (Zymo Research). Libraries were prepared on V3-V5 region of 16S gene and sequenced on Miseq with PE 300bp output. In addition, samples of Total Mixed Ration (TMR) were collected and analysed. Bioinformatic analysis was performed using QIIME2 pipeline. Sequences were clustered into Amplicon Sequence Variant (ASVs) using DEBLUR, and the taxonomic assignment was conducted using the GreenGenes2 database. Statistical analysis (i.e. β -diversity metrics and identification of differentially abundant species) was done with DESeq2. Results about TMR composition show high variability in the rations. Considering a different forage-concentrate ratio (F:C) from high ($\geq 80:20$) to low ($\leq 70:30$), the metagenomics preliminary results highlight some significant differences. In rations with low F:C there is a greater abundance of amylolytic and lipolytic species (i.e. *Clostridium bormimense* and *Anaerovibrio lipolyticus*). In conclusion, ration composition influences the microbial communities also in buffalo faeces.

Assessment of infrared spectroscopy for the quantification of protein fractions in buffalo milk

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Despite the subject of cow milk proteins have been extensively addressed, there still is a lack of studies investigating detailed protein profile in buffalo milk. The aim of this study was to qualitatively and quantitatively determine protein fractions in buffalo milk. An additional aim was to assess the effectiveness of mid-infrared spectroscopy to predict milk protein composition. Individual milk samples of Italian Mediterranean buffaloes (n = 401) were collected in 7 commercial herds located in the area of Mozzarella di Bufala Campana PDO (south of Italy). Traditional milk quality traits (fat, protein, casein and lactose, %), pH and urea content (mg/dL) and spectral data were obtained using MilkoScan FT6000 (Foss Electric A/S, Hillerød, Denmark). Detailed milk protein composition (α S1-, α S2-, β -, and κ -casein, α -lactalbumin and β -lactoglobulin) were determined by reversed-phase HPLC. Within sample, the concentration of each protein fraction was matched with the corresponding spectrum. Chemometric analyses were carried out using WinISI 4 software (Infrasoft International, Port Matilda, PA) applying a 5-fold cross-validation. Results showed moderate accuracies of the mid-infrared spectroscopy to predict different protein fractions, with a coefficient of determination in cross validation (R^2) from 0.34 (β -lactoglobulin) to 0.60 (β -casein). In perspective, further research should be addressed to improve the accuracy of near-infrared spectroscopy models by increasing the number of samples in calibration, or considering alternative chemometric approaches.

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Biosecurity correlation with Antimicrobial Use and Animal Welfare in buffalo farm evaluated by ClassyFarm System

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Biosecurity in ruminant production is often an overlooked area, instead, it is a key aspect of the interactions among the animal production systems, the environment, and human health in a One-Health approach. ClassyFarm is an integrated system for categorising farms according to the risk assessment methodology. ClassyFarm gathers and processes data related to biosecurity, animal welfare (AW), and antimicrobial usage (AMU). It can be applied to several livestock species, including water buffalo, for which biosecurity is evaluated on 15 items. The assessment of AW includes 63 non-animal-based measures (N-ABMs) and 17 animal-based measures (ABMs). The N-ABMs are divided into 'Management' (32 items) and 'Housing' (31 items). Total and partial scores are expressed in percentages. AMU was estimated using the Defined Daily Animal Dose for Italy (DDDAit). The Biosecurity & AW were assessed on 382 buffalo farms, with an average size of 374 heads. The mean overall AW score was $76.9 \pm 9.8\%$ and the Biosecurity was $68.9 \pm 15.3\%$ and DDDAit 0.40. Statistical analysis was performed by Spearman's rank correlation using GraphPad Prism 8.0.1. Biosecurity and AW were positively correlated ($\rho=0.42$; $p < 0.001$). A positive correlation was observed between Biosecurity and both 'management' ($\rho=0.61$; $p < 0.001$) and 'housing' ($\rho=0.36$; $p < 0.001$). No correlation was found between AMU, Biosecurity and AW respectively, likely due to the low levels of AMU found. The low AMU values confirm the peculiarity of buffalo breeding, a strong point to be exploited in communication. Knowing the Biosecurity and AW levels, their relationship and critical point in animal farms, represent a tool to plan the strategic priorities of human resources and investments, to increase the Farmers' awareness and the health, and ethical sustainability of the farm.

A new version of buffalo SNP array to support Mediterranean breed genomic improvement

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Domestic water buffalo is an important worldwide species, despite the smaller population in comparison with bovine. Two types of buffalo are recognized: swamp and river. The former are populations supporting people in many developing countries for draft purposes. The latter are breeds mainly selected for milk production. In Italy, Mediterranean (MED) river breed is bred, from which the famous “Mozzarella di Bufala Campana” is obtained. Recently, the breeding association ANASB published the first genomic breeding index for MED. However, the use of the current buffalo array (ABA1) has limitations (eg, number of usable markers and their density), because its aim was for biodiversity characterization and not genomic selection in MED. In the framework of the GENO-BU project, thus, a specific MED version of the Axiom Buffalo array was developed. The markers included in the ABA1 were validated using previously genotyped MED animals; compatibility with the ABA1 and IMAGE array was considered, and SNPs associated with milk characteristics were prioritized. Finally, 72,223 SNPs were retained. New markers were identified using short and long reads (ONT) sequences from MED animals (unpublished data kindly provided by F. Salvatore and G. Paoletta, CEINGE, Napoli, Italy, within the GENOBU project), to optimize the array density and fill the almost 123k spots available. The data were aligned to the most recent MED reference sequence to identify new variants, using state-of-the-art bioinformatic pipelines. Markers from Y chromosome, together with SNPs from candidate genes affecting functional traits (eg, casein cluster), were also included. The final number of markers included in the new version is 122,646, reaching 21.8±7.7 kbp average marker distance.

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Influence of udder type traits on stayability in the Italian Mediterranean buffalo

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In previous studies, stayability has been found to be a useful tool to assess longevity in Mediterranean Italian Buffalo. Animal longevity has been shown to correlate with type traits in dairy cows and in dairy buffaloes, as long as milk production increases thanks to genetic selection. In buffaloes, morphology was found to influence stayability up to parity 7, after which milk production was the discriminating trait. A wide thorax, properly angulated and with a good diameter, together with a longer trunk, allows higher production without compromising the health status of the animal, leading to greater longevity. A correct rump conformation, wide in both fore and hindquarters and only slightly angulated, also allows greater longevity, probably due to its involvement in reproductive events. The aim of this study was to correlate longevity with the udder traits measured by ANASB for IMB. The ANASB dataset including 49,667 animals born between 2005 and 2018, with type trait evaluations carried out between 2010 and 2021 was used. Data preparation and all statistical analyses were performed using the R programming environment v.4.3.0 (R Core Team, 2023). The R package lme4 (Bates D, 2015) was used to run a mixed effects logistic regression model using the 'glmer' function, including milk yield, all type traits for udder conformation included in the ANASB type trait evaluation, age at first calving as fixed effects, and the interaction of herd-year of evaluation-inspector as a random effect. The morphological traits that were found to have a significant influence on stayability, especially at parity 1 to 5, were mainly udder depth with an inverse relationship and mean ligament strength with a positive relationship. However, milk production was always the most important trait for culling. Acknowledgements: This study was supported by 10.2 of the National Rural Development Programme (PSRN)-Biodiversity 2014–2020, BIG project Prot. N. 0215513 CUP ANASB: J29J21003720005.

How old is too old? Effects of parity in dairy water-buffalo on milk and reproduction efficiency

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Longevity of dairy water-buffaloes is not well documented to-date, with some reports mentioning that cows remain fertile and are able to sustain new lactations even after 18 years of age. The aim of the current research was to evaluate the effects that parity has on production and reproduction performances in dairy water-buffalo cows. Therefore, a study-herd of 56 Romanian buffalo breed multiparous cows were monitored at the Research and Development Station for Buffalo Sercaia, with an average parity of 6.42 ± 0.40 , ranging between 3rd and 15th. Three age groups were set-up, as follows: YOU – parities III-V ($n=24$), MID – parities VI-VIII ($n=20$), and OLD – parities IX and above ($n=12$). Data on milk yield during the first 100 days of lactation (100DIM), milking ejection rate (MER), body condition score (BCS) and calving intervals (CI) were evaluated using the Mann Whitney U non-parametric test. Milk yield varied between 788.0 ± 119.0 and 882.0 ± 105.0 kg/100DIM among groups, with no effects ($p > 0.05$) of the parity. Similarly, MER and BCS were not influenced ($p > 0.05$) by the parity of the buffalo cows, with averages ranging between 0.43 ± 0.07 and 0.52 ± 0.08 kg/min and between 6.2 ± 0.34 and 7.0 ± 0.26 , for MER and BCS, respectively. Moreover, contrary to our expectations, the YOU group had an average CI of 535.2 ± 78.6 days, compared to the shorter CI intervals of 476.1 ± 69.1 and 471.8 ± 47.3 days, for the MID and OLD group, respectively. Current results outline that older water-buffalo cows maintain both appropriate fertility and productivity throughout latter parities.

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Poster 14

Accuracy of a fast low-priced device based on 2D image analysis in predicting body weight of Italian Mediterranean buffalo

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Body weight (BW) is important in the productive and reproductive management of the Italian Mediterranean buffalo (IMB). Identifying anomalous variations promptly allows for the early detection of metabolic disorders or deficiencies. Traditional method implies human labor and is time consuming and stressful for animals. BW can be also measured by image analysis (IA). The present study was performed to evaluate the precision of a rapid tool, based on IA, in BW prediction of IMB when compared to official method. Fifty-five lactating buffaloes (first to sixth parity) were weighed using a livestock scale in the morning, prior to feeding. Meanwhile, using a mobile device, equipped with a laser telemeter to carry out continuous measurements from a distance, a body lateral image was acquired and processed by an App for BW assessment. The surface area of all lateral profile was calculated and used in predicting BW using a previously developed equation (modified). High correlation ($r=0.95$ $p < 0.0001$) was found between predicted and measured BW. Regression App prediction on measured BW ($y = 0.98x + 16.71$ $R^2 = 0.90$, $p < 0.0001$) showed a slope coefficient very close to one and an average measurement error of 2.5% (RMSE = 18.45). Residuals analysis did not show any systematic error. In conclusion a rapid and accurate measure of morphometric traits for BW prediction may be easily obtained either by generic operators or automatized systems, during the routine operations.

Analysis to the microbiome of buffalo milk and mozzarella cheese using next generation sequencing techniques

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Milk and dairy products are a fundamental component of the human diet due to their digestibility and high nutritional value. In recent years, the development of molecular techniques, such as Next-Generation Sequencing (NGS), has dramatically increased our knowledge of the composition and structure of the microbiomes of raw milk and milk-derived products. The Amaseno (FR) valley is known for producing mozzarella and typical cheeses from buffalo milk. The metagenomics analysis carried out on the buffalo milk samples from three dairy farms for one year showed that the structure and composition of the buffalo milk microbiome were strongly influenced by farm management and seasonal trends (temperature and humidity). The analysis also revealed the presence of lactic acid bacteria with technological and beneficial traits belonging to the genera *Lactococcus*, *Streptococcus*, and *Lactobacillus*. The NGS analysis on mozzarella samples showed an increased relative abundance of *Streptococcus thermophilus* and *Lactobacillus crispatus*-affiliated taxa. *S. thermophilus* could play an essential role in the cheese-making process by providing peculiar sensory properties due to the production of aromatic compounds. At the same time, *L. crispatus* might have potential probiotic activities. Our data indicate that microbiome analysis is a valuable tool to characterize the milk and mozzarella's microbial community by highlighting the presence of taxa with dairy properties that benefit human health.

Effect of seasonality and farm management on the buffalo's milk lipidome

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The milk characterization using untargeted lipidomics based on mass spectrometry techniques can provide valuable information on the total lipid profile, the temporal variations in the composition and characteristics of the lipid fraction, and the association between these variations and environmental factors. This study evaluated the association between different feeding management and seasons to bulk milk lipidome in 8 buffalo farms from the same geographical area (Amaseno, Italy). Milk samples were collected from January to November and amended with 2-bromo-2-nitropropane-1,3-diol as a biocide and preservative. The data analysis allowed us to identify biomarkers that helped evaluate the impact of seasonality and farm management on lipidome. Different lipid extraction protocols were used, including liquid-liquid extraction and solid phase extraction, showing how the result, in terms of the number of detectable chemical fingerprints, was strongly affected by sample preparation. With the optimized protocol, the buffalo milk lipidome was shown to consist of approximately 515 compounds (10% greater than that of the bovine milk lipidome). The chemometric analysis demonstrated that the macroscopic differences among the buffalo milk samples resulted from 8 compounds belonging to 7 distinct classes. In contrast, the micro-differences among milk samples referred to a group of 16 compounds, 12 of which belong to the class of chain triglycerides between C8 and C20.

Influence of breeding techniques during the dry period on welfare in buffalo species

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The aim of the study was to evaluate the influence of two rearing techniques on welfare status of Italian Mediterranean buffaloes (IMB) during the dry period. Twenty IMB were selected and shared in two groups according to parity and milk yield (MY): Group P was kept under extensive conditions and group S in barn. Blood samples for biochemical analyses and hair samples for cortisol measurement were collected monthly, from April to September. In addition, daily temperature and relative humidity were recorded to calculate the Temperature Humidity index (THI). At the end of the dry period, individual MY and milk quality were evaluated monthly. Significant differences were observed between group P and S for total protein (8.19 and 7.52 g/dl, $P < 0.01$), triglycerides (20.77 and 28.75 mg/dl, $P < 0.01$) and urea (45.89 and 38.43 mg/dl, $P < 0.05$). Cortisol concentrations tended to differ ($p < 0.10$) between group P and group S (7.73 and 4.51 pg/mg, respectively). Group P showed a higher milk yield compared to group S ((11.19 vs 10.64 kg/die, $p < 0.01$), and lower fat concentration (7.80 vs 8.37 %, $p < 0.05$). THI was negatively correlated to triglycerides and cholesterol (-0.36 and -0.39 respectively, $p < 0.001$), and positively to cortisol and LDH (0.38 and 0.33 respectively, $p < 0.001$). In conclusion, providing physical exercise during the dry period seems to have a positive influence of on fitness and MY during the subsequent lactation, so more research is needed possibly recruiting a larger number of buffaloes.

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Poster 18

Meat and milk from dual-purpose water buffalo production systems in Greece

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The objective was to assess the characteristics and the potential of dual-purpose water buffalo (*Bubalus bubalis*) farms in Greece towards production of high-quality meat and milk. We provide suggestions and objective solutions to ensure farm sustainability emphasizing animal health, welfare, and product marketability. Phenotypic data are collected from a representative number of animals. Meat quality is assessed through pH, moisture, fat and protein content, fatty acid profile, color, water holding capacity, and morphometric analysis of skeletal muscles. Milk quality is assessed through microbiological and physicochemical analyses. Based on the latest census (2023), the national herd comprised 6,740 animals kept on 38 farms; an increase of 53% compared to 2014. Age and sex structures comprise 589 females and 436 males <6 months, 979 females and 831 males between 6-24 months and 3,367 females and 538 males >24 months old. The focus of farms is meat production. Over the last 3 years, the average number of animals slaughtered was 838, aged between 18-24 months, and produced carcasses of 220-250 Kgs. About 18% of farms also produce milk sold to local dairies or used for homemade dairy products. The average lactation period is 150±10 days. About 20% of the breeders apply an intensive rearing system for the animals sent to slaughter. Preliminary results provide useful insights into the meat and milk quality produced by existing buffalo farms in Greece. This work is funded by Bubalis project (Measure 16:Cooperation, Project code M16ΣYN2-00193).

The use of coproducts to mitigate the environmental footprint of pigs and broiler chicken production

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Mitigating livestock's environmental footprint is an important pillar for building sustainable animal production. This could be done by improving feed efficiency thanks to the use of feed additives. This study aimed to assess the environmental impacts of the use of a commercial citrus extract feed additive (CEFA) in swine and broiler chicken farming. Life-cycle assessment (LCA) was applied to assess the impact of manufacturing and distributing one 25 kg bag of CEFA and its use in broiler and swine productions. Results showed that most of the impact of CEFA manufacturing came from the production of CEFA ingredients, accounting for 70% of the impact generated. The remaining 30% effect was divided between transportation to the customer (25%), CEFA packaging (3%), and CEFA manufacturing and production loss (2%). Moreover, the use of CEFA in pigs and broiler production was shown to improve environmental indicators such as climate change, land occupation, and water consumption, compared to standard conditions without CEFA. Indeed, CEFA supplementation allowed to enhance growth performances, hence reducing the required amount of consumed feed to achieve the same level of growth. Consequently, this helped reduce environmental issues from animal feed ingredients' agriculture. To be more specific, the use of one 25 kg bag of CEFA led to a reduction of 6 tons of CO₂ equivalent emitted along the life cycle of poultry production and 5 tons in the case of fattening pigs. The inclusion of CEFA in the diet also led to a reduction in the land use footprint by 0.7 hectares and reductions in water consumption by 201 m³ and 82 m³ for broiler and swine production, respectively. The LCA thus showed the interest in using this CEFA in swine and broiler chicken diets to mitigate the environmental impacts.

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Theatre 2

Methane emissions from beef fed different protein sources and a forage-based diet

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This study assessed the effect of different dietary protein sources (soyabean, SB; brewers' spent grains, BSG; local field beans, BNS) on methane (CH₄) emissions of beef cattle fed a 70:30 forage:concentrate basal diet, and also compared with a 100% pasture-fed beef system (PAS). 16 Holstein × Angus growing steers were allocated to four experimental diets (SB, BSG, BNS, PAS; four animals each) for 16 weeks. Every 4 weeks animals were transferred in digestibility stalls (for nutrient balance measurements) within respiration chambers (for CH₄ emission measurements) for 4 days. During the chamber measurement periods, dry matter intake (DMI, kg/day) and gross energy intake (GEI, MJ/day) did not differ between groups (P>0.05). PAS steers had lower CH₄ production (g/day; P<0.001) and CH₄ yield (g/kg DMI, P=0.001; MJ/MJ GEI, P=0.005) compared with the three concentrate-fed groups. Faeces and urine energy outputs (expressed as % GEI) were higher (P<0.001) in PAS steers compared to the concentrate-fed beef. There were no significant differences (P>0.05) in the studied parameters between the three concentrate-fed groups. The lower CH₄ production and yield in pasture-fed steers could indicate potential environmental benefits associated with pasture-based beef systems, but the higher energy loss in faeces and urine demonstrates potential inefficiencies in energy utilisation.

Effect of dietary protein concentrations on feeding efficiency and methane emissions of dairy cows across lactation

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This study assessed the impact of dietary CP level (g/kg DM: Low, LCP, 122; Medium, MCP, 151; High, HCP, 181) on productivity, feeding efficiency and methane production (CH₄) of Holstein-Friesian cows across lactation. 12 primiparous and 12 multiparous cows were allocated to 3 diets (LCP, MCP, HCP) in a randomised blocked design throughout lactation (1–305d). During early (70–90d), mid (150–170d) and late (230–250d) lactation, 6-day digestibility measurements were performed in metabolism units, followed by 3-day gas exchange measurements in respiration chambers. Data was analysed by a linear mixed model with repeated measures using CP level (LCP, MCP, HCP), stage of lactation (early, mid, late), parity (primiparous, multiparous) and their interactions as fixed effects. Regression equations were developed to identify relationships between diet CP content and measured variables. MCP and HCP cows had higher energy-corrected milk yield (ECMY; 25.7 and 27.8 kg/day), ECMY/dry matter intake (DMI) (1.40 and 1.42 kg/kg) and milk energy/DMI (4.33 and 4.39 MJ/kg DM) than LCP cows. Regression analysis showed that milk yield was quadratically increased beyond 150 g CP/kg DM, with a peak shown at 170g CP/kg DM, but this also depends on lactation stage. The HCP group had higher energy digestibility (0.74 MJ/MJ) than the LCP (0.72 MJ/MJ) and MCP (0.72 MJ/MJ). CH₄/ECMY was higher for LCP (17.9) than MCP (14.8) and HCP (14.5) cows.

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Theatre 4

Dietary crude protein reduction in poultry diets allows to decarbonize poultry meat and egg production with different levels of expected benefits across segments

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Feed accounts for around 75 % of the environmental impact of the poultry meat and egg production, regardless of the production method. This study aimed to compile recently conducted poultry experiments with the same goal and methodology: studying the impact of reduced dietary crude protein (CP) levels on growth and laying performance as well as measuring the benefits in terms of carbon footprint. Five different trials were compiled and compared across broiler genetic lines (Ross 308, JA 787, S757N) and segments (broiler, turkey, duck, and brown-laying hen). Dietary CP reduction was achieved in all segments by gradually replacing imported soybean meal by cereals, and feed-grade amino acids (AA) controlled at least at the assumed requirement. Dietary digestible Lysine (dLys) supply was constant for all treatments within each experiment. With this protocol, it was possible to reduce dietary CP in diet formulation, expressed as mg dLys/ point of dietary CP as low as: 55 to 63, 49 to 58, 45 to 54, 47 to 57, 56 to 61, and 41 to 50, for Ross 308 broiler, JA 787 broiler, S757N broiler, duck, turkey, and laying hen, respectively. Based on growth performance, meat yields and egg performance, the lowest reachable levels of dietary CP reduction, without impacting performance were: 63, 52, 45, 56, 45 mg dLys/pt CP for Ross 308, JA 787, S757N, turkey and laying hen respectively. It is possible to reduce dietary CP in all segments and species, but the level of nutrient density reachable differed within the broiler segment, and between species. By reducing dietary CP, reducing imported soybean meal and implementing MatriCIEL® calculations, it was also possible to reduce carbon footprint (in kg CO₂eq/kg meat or egg) by 2-15 % for Ross 308 broilers and by 4-10 % for layers.

Effect of fresh hemp and savory leaves on feed intake and rumen fermentation: in vitro and in vivo trials

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The pharmaceutical and herbal sectors produce large amounts of by-products, potentially usable in dairy cows' nourishment. In this contest, fresh hemp leaves (*Cannabis sativa* L.; HL) and savory leaves (*Satureja hortensis* L.; SL) were tested in vitro and in vivo to evaluate their impact on rumen fermentations. As first step, HL and SL fermentation properties were compared with grass hay using an in vitro gas production (GP) system. Then, 6 lactating cows in a 3×3 Latin Square design were fed in vivo with HL and SL with a dosage of 3 kg DM/d to evaluate the impact on feeding behaviour and rumen fermentations. Finally, the microbial activity of individual rumen fluids (RF), collected at the end of each period of the in vivo trial, was evaluated again in vitro. All RF samples were analysed for pH, volatile fatty acid (VFA), and ammonia. For the in vitro trials, GP kinetics and gas composition were also measured. First, in vitro results showed that, compared to grass hay, HL exhibited a higher degradability, a slower GP kinetic and an increased acetic+butyric/propionic (ABP) ratio, CH₄ production (mL/g DM) and intensity (% v/v). Conversely, SL showed a lower degradability and GP, a higher ABP ratio and a lower CH₄ production and intensity. For the in vivo and the later in vitro trials, HL reduced the feed intake and slightly increased propionic and valeric acids in the RF, whereas SL was favourably accepted by the cows but did not affect rumen fermentations. Our findings highlighted the feasibility of utilizing these herbs in ruminant feeding, but further investigations are required to assess the impact of higher quantities, particularly on long-term intake. Research funded by the projects: UNIPD 2021, BIRD213117

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Theatre 6

Effect of β-mannanase on growing-finishing pig performance, economics, and carbon footprint

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It is speculated that β-mannanase (MAN) can improve nutrient efficacy, though studies that evaluated the efficacy of MAN on performance of growing-finishing (GF) pigs didn't reduce the energy (E) content of the experimental diets to test this concept. The objective of this experiment was to evaluate the effect of MAN (Hemicell™ XT; Elanco AH, Indianapolis, IN) on performance, carcass characteristics, economic viability (IOFC), and carbon footprint in GF pigs fed a diet lower in NE (-35 kcal NE/kg). It was hypothesized that treatments (T) would perform similarly. The trial consisted of T1 the control diet (CD) and T2) the CD + MAN (133 g/ton). A starter diet was fed between 25-50 kg (CP 16.8%, SID Lys 10.3 g/kg, T1: NE-value 2450 or T2: 2415 kcal/kg), a grower diet between 50-85 kg (CP 14.7%, SID Lys 8.65 g/kg, T1: 2400 or T2: 2365 kcal/kg), and the finisher diet between 85-120 kg (CP 13.4%, SID Lys 7.50 g/kg, T1: 2350 or T2: 2315 kcal/kg). The diets contained palm kernel expeller and soybean hulls to achieve a β-mannans content commonly present in EU diets. The difference in NE was created by the removal of animal fat (-34%). Each T had 12 replicates consisting of 8 GF (male:female ratio 1:1). Pigs weighed 25.4 kg (SD=1.27) at d0. Pigs were individual weighed at each diet transition and the day before slaughter. Feed intake was recorded during each phase. Treatment did not significantly influence pig performance or carcass characteristics (P>0.05). T2 resulted in a profit of €0.49 per pig and could reduce the carbon footprint by ≥1.00%. The results indicate that MAN whilst in a diet with a lower E content does not negatively influence performance and may result in a lower carbon footprint and reduced production costs.

Exploring Kaempferol 3-O-Glucoside as a Potential Feed Additive: In vitro Assessment for Mitigating Methane Emissions in Livestock Nutrition

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The aim of this study was to evaluate by in vitro gas production technique the supplementation of pure kaempferol 3-O-glucoside, a metabolite abundant in the leaves of *Fagus sylvatica* L. Different dose levels of pure compound (0, 5, 50 and, 500 µM/g as feed) supplemented to a standard diet were tested in vitro with buffalo rumen liquor evaluating gas production (OMCV), organic matter digestibility (OMD) and fermentative end-products (pH, volatile fatty acids, VFA) after 120 h of incubation. Effects on methanogenesis, organic digestibility and volatile fatty acids production were evaluated after 24 h of incubation. Fermentation liquor after 0, 24 and 120 h of incubation was analysed through UHPLC-ESI-QqTOF-MS/MS analysis to follow the molecule biotransformation. The polynomial contrasts were used to observe differences between the doses (0, 5, 50, and 500 µM/g). Among the different tested dose levels, 50 µM/g resulted as optimal in terms of OMD, OMCV with a reduction of methane production after 24 h of incubation. The supplementation of 50 µM/g did not significantly modify OMD after 120 h, providing a significative increase of acetate and valerate. The fermentation kinetics significantly increase at 5 and 50 µM after 120h. The metabolomic analysis of the compound in fermentation fluid suggested the occurrence of rumen biotransformation during incubation period. In conclusion, kaempferol 3-O-glucoside inclusion in livestock diets modulates ruminal fermentations, suggesting its potential as a feed additive for methane mitigation.

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Theatre 8

Quantifying methane emissions and animal performance from zero-grazed beef steers supplemented with *Ascophyllum nodosum* via a water-based delivery system

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Quantifying baseline methane emissions from grass-fed beef production is vital in generating an accurate Irish agricultural inventory. In addition, identifying a suitable strategy to administer a methane mitigating additive at pasture is crucial in reducing agricultural emissions. Therefore, this study aimed to quantify methane emissions from grass-fed steers and examine the impact of *Ascophyllum nodosum* seaweed extracts delivered via a water-based delivery system. Forty-five beef steers were assigned to three dietary treatments in a complete randomized block design. Diets, comprising of zero-grazed grass offered ad-lib once a day, included either no seaweed (CTR), 1% of dry matter intake (DMI) of seaweed 1 (SW1) or 4% DMI of seaweed 2 (SW2) delivered through the water. Methane was continuously measured using GreenFeed (GEM) with DMI measured in weeks 5 and 10. GEM visits per animal averaged 240 with a mean 4.44 visits/d and duration of 04:37 mins/visit. Steers had a DMI of 7.34 ± 0.83 kg/d, average daily gain (ADG) of 1.46 ± 0.28 kg/d and methane output of 207.32 ± 17.52 g/d, methane yield of 29.11 ± 2.86 g/DMI and methane intensity of 146.74 ± 26.22 g/ADG. Seaweed supplementation had no effect on any measured parameters (P >0.05). To conclude, the methane emissions and performance of beef steers offered zero-grazed grass was quantified with no effect of seaweed supplementation. This study identifies the potential for water-based delivery systems to provide methane mitigating additives in pasture-based systems.

Effect of rape seed fat and saturated fat on milk production and enteric methane, when added to diets with different ratios between grass-clover and maize silage

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The study aimed to assess the impact of supplementing rape seed fat or saturated fat to diets with different grass-clover:maize silage ratios on enteric methane (CH₄), and milk production. The design was a 4×4 Latin square with 4 treatments and 4 Holstein cows over 4 periods of 21 days. The last 4 days of each period dry matter intake (DMI), milk yield, milk composition, and enteric CH₄ were measured in respiration chambers. All diets comprised 59% silage DM, with 2 silage ratios (either 80:20 or 20:80 grass-clover: maize silage), and either rape seed fat or saturated fat (45:45:10 palmitic:stearic:oleic acid), resulting in 51 g fatty acids (FA)/kg DM. Starch content was higher in high maize diets (249 g/kg DM) than in low maize diets (183 g/kg DM). No significant interactions were found. Significant effects were observed for silage ratio and fat source on DMI, milk fat% and Energy Corrected Milk (ECM). High maize increased DMI, reduced CH₄/kg DMI, lowered milk fat%, and resulted in the lowest ECM yield. Rape seed fat led to reduced DMI, lowered milk fat%, and the lower ECM yield. Due to reduced milk fat%, a combination of high maize silage and 51 g FA/kg DM partly from rape seed does not seem to be a feasible CH₄ mitigation strategy, despite the lowered methane yield.

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Mango and Avocado Byproducts as Feed Ingredients and Additives in Ruminant diet

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The intensification of food-feed competitions and environmental concerns has driven research into unconventional feed sources for optimizing ruminant feeding. In this study, we explored the potential use of mango peel (MP), mango seed (MS), mango seed coat (MSC), avocado peel (AP), and avocado seed (AS) in two experiments. Experiment 1 evaluated the feed potential of these fruits byproducts by assessing their chemical composition, in vitro true digestibility, gas production, and volatile fatty acid production. In vitro true digestibility was determined using the Ankom DaisyII incubator. Among the various byproducts, MP and AP exhibited higher total phenolic content ranging from 121.50 to 243.69 (mg GAE/g) and antioxidant capacity from 342.92 to 366.63 (mg TE/g), indicating their potential to positively influence the rumen ecosystem. MP, MS, and AS showed higher digestibility (86.4–89.5%), increased gas production, and elevated metabolizable energy (8.41–9.59 MJ/kg DM), while MSC and AP exhibited lower values. Acetate-to-propionate ratio, a key methane indicator, was higher in AS (3.97), MSC (3.76), and MS (3.53), and lower in AP and MP (3.05 and 3.08, mmol/l respectively). Experiment 2 was conducted to assess the effectiveness of incorporating mango and avocado by-products extracts (20 mg) as feed additives, in comparison to alfalfa hay utilized as a control basal diet (200mg DM) across all groups. The findings demonstrated that the AS extract group exhibited the highest 24-hour gas production (47.3 ml/0.2 g DM) when compared to both the control and other extract groups (42.1 to 42.5 ml/0.2 g DM). MP and AP extracts significantly reduced acetate to propionate ratio compared to control and other extracts ($P < 0.001$). Furthermore, the extract groups exhibited a lower ammonia nitrogen concentration compared to the control. In conclusion, MP, MS, and AS show promising results as feed ingredients. Peel extracts of mango and avocado emerge as feed additives, modulating rumen fermentation parameters effectively.

Effect of *Cymbopogon citratus* on Enteric Methane Emission and Energy Partitioning in Growing Beef Cattle
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Globally, cattle production systems dominate the agricultural sector CH₄ emissions with 64-78% (2.3 to 2.8 Gt CO₂ eq-1). Therefore, it is necessary to develop natural and sustainable CH₄ mitigation strategies for cattle production systems, particularly for developing countries. Evidence suggests lemongrass, *Cymbopogon citratus* (CC), can reduce enteric CH₄ production. The objective of the present study was to evaluate the anti-methanogenic effect of increasing the supplementation levels of CC on the dry matter intake (DMI), digestibility, CH₄ production, and partitioning of the gross energy intake in growing beef heifers fed with a diet high in forage (68.6% forage: 31.4% concentrate). An experiment was conducted using Holstein × Charolais heifers distributed in a 4 × 4 Latin square design. The experimental treatments were: (1) control diet (CO), (2) CO + 30 g CC DM/d, (3) CO + 60 g CC DM/d, and (4) CO + 90 g CC DM/d. A reduction of 22.4% in methane yield (CH₄ g/kg DMI) and a reduction of 21.2% in the Y_m factor was observed with the 30 CC treatment ($p \leq 0.05$). However, no significant differences ($P > 0.05$) were observed for the total daily CH₄ production, DMI, nutrient digestibility, and gross energy intake partitioning in the heifers. Therefore, we concluded that the 30 g CC DM/d supplementation reduced the CH₄ yield without affecting the animal performance. However, the anti-methanogenic properties of *Cymbopogon citratus* deserve more investigation.

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Theatre 12

Including dried algae in dairy cow diets decreases emissions, yield and intensity of enteric methane
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This study assessed the effects of supplementing dairy cow's diet with dried algae on enteric CH₄ emissions and milk production of Holstein cows. Sixteen multiparous lactating cows (DIM = 162 ± 29.5) were used in a completely randomised block design. The experimental period consisted of 5 weeks preceded with 3 weeks of covariate. Cows were fed (ad libitum) a total mixed ration not supplemented (Control), or supplemented (0.25% of dry matter) with dried algae (Synergraze Inc., Calgary, Alberta, Canada). The dried algae contained 4.65 mg/g bromoform. Intake of DM (DMI) and CH₄ production (4 days; respiration chambers) were determined during the covariate period, at the beginning (week 1), and after 35 days (week 5) of feeding the algae whereas, milk production and milk were determined weekly (including covariate period). Data were analysed (MIXED Procedure, SAS) using the repeated statement adjusted for the covariate. Significant effect was declared at $P \leq 0.05$. The interaction between the experimental treatment and sampling week was not significant for CH₄ production, DMI, or milk production. Algae supplementation had no effect on DMI (25.9 kg/d), milk production (39.3 kg/d), or milk fat (3.79%) and milk protein (3.36%) percentages. In contrast, compared with the control, feeding algae reduced ($P < 0.01$) enteric CH₄ emissions (g/d) by 45%. Consequently, methane yield (g CH₄/kg DMI) decreased from 19.3 g/kg to 10.6 g/kg and CH₄ emission intensity (g CH₄/kg energy-corrected milk) from 12.47 g/kg to 6.53 g/kg upon diet supplementation with algae. This study shows that including algae at 0.25% of DMI caused a marked reduction in CH₄ yield and CH₄ emission intensity by 45% and 48% respectively, without adversely affecting DMI or milk yield.

Phenotypic traits related to methane yield in dairy cows

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Dairy cows vary in CH₄ yield (CH₄ per kg of dry matter intake (DMI)) and the trait has some heritability. Knowledge of the phenotypic traits related to variation in CH₄ yield is essential for proper use of this variation to decrease methane emission from dairy production e.g. by breeding. Cows (16 Danish Holstein) were surgically fitted with a rumen cannula in connection with the drying off after 1st, for use in 2nd lactation. The experiment included digesta sampling for determination of nutrient digestibility, rumen fermentation and microbiome pattern, rumen evacuations for rumen pool size and fiber kinetics, and methane measures in respiration chambers. A 2×2 cross-over design with 2 treatments and 2 periods of 28 days was used. Treatments were diets with either 35% or 63% forage. Forage consisted of 80% grass-clover silage and 20% maize silage, and concentrate of 61% barley, 30% rapeseed meal, and 9% dried beet pulp, all on DM basis. Mineral and vitamin supplements were the same across diets. The cross-over design allowed extraction of the random cow effects (RCE) using a mixed model in R. The RCE of measured variables were related to RCE for methane yield using simple linear regression and Pearson correlations. Methane yield RCE (g/kg DMI) was positively correlated to RCE for organic matter digestibility ($r = 0.64$, $P < 0.01$), RCE for rumen ventral concentration of acetic acid ($r = 0.56$, $P = 0.02$) and RCE for rumen relative abundance of the methanogen *Methanomassilicoccus* ($r = 0.52$, $P = 0.04$), but not correlated to rumen pool size. Breeding for cows with a lower CH₄ yield might render cows less efficient in digesting feed, unless also digestibility and efficiency measures are included in the breeding program.

Session 34

Poster 14

Effects of high-fat oats, rapeseed cake, and 3-NOP on milk production and methane emissions of dairy cows

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The objective was to examine the effects of high-fat oats (HF-oats), cold-pressed rapeseed cake (cpRSC), and 3-nitrooxypropanol (3NOP) on milk production and CH₄ emissions in Nordic Red dairy cows. We enrolled 24 lactating cows, divided into blocks, in a cyclic change-over design replicated in 4 periods of 28 days each. Diets comprised barley or the HF-oats variety Fatima as energy source and rapeseed meal (RSM) or cpRSC as protein source. Diets were fed with or without 3NOP supplementation at 60 mg/kg DMI. The forage to concentrate ratio was 60:40 with grass silage as sole forage. Gas emissions were measured by the GreenFeed system. Data were analyzed in SAS by the MIXED procedure as a 2×2×2 factorial design. Dietary treatment did not affect DMI. Milk and ECM yields were higher ($P < 0.01$) when feeding HF-oats instead of barley (41.3 vs. 38.9 kg ECM/d, respectively). Supplementation with 3NOP decreased ($P = 0.02$) milk yield by 1.2 kg/d, but did not affect ECM. We observed an interaction effect ($P = 0.05$) of energy and protein source on CH₄ yield (g/kg DMI) and CH₄ intensity. Feeding HF-oats + cpRSC decreased CH₄ intensity by 8.5% compared with barley + cpRSC (7.6 vs. 8.3 g/kg ECM, respectively), but feeding HF-oats + RSM decreased CH₄ intensity by 15.0% compared with barley + RSM (8.0 vs. 9.4 g/kg ECM, respectively). Supplementation with 3NOP decreased ($P < 0.01$) CH₄ intensity by 31.3%. In conclusion, combining HF-oats, cpRSC, and 3NOP mitigates enteric CH₄ emissions, but the effects are not additive.

Repeatability of enteric methane measurements using GreenFeed systems on upland pastures

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Plant diversity of mountain pasture has been shown to reduce enteric methane (CH₄) production in vitro. In 2023, we set up an in vivo experiment to measure enteric CH₄ emission using GreenFeed (GF) systems and dairy performances of 28 grazing dairy cows on mountain pasture. After 3 weeks of being fed fresh grass indoor, they were taken out to extensively managed pasture for 9 weeks. Cows were allowed to graze large areas (from 1 to 3.5 ha), and 2 GF equipped with solar panels were accessible near the water trough. As lower frequency of animal visits to the GF at pasture than indoor was expected, our aim was to determine the minimum number of consecutive CH₄ spot measurements (CSM) to achieve a good repeatability of CH₄ data. As expected, the average number of visits per animal and per day (d) was 2.6 ± 0.42 vs 0.7 ± 0.24 during indoor vs grazing periods. Five to 20 CSM were randomly chosen within the set of available data per animal per period (indoor vs outdoor) and the draws were repeated 10 times. The analysis was repeated 5 times to evaluate the consistency of the results across draws. Results showed that a minimum of 15 CSM acquired in 12 and 29 d was necessary to achieve a similar repeatability of 0.53 and 0.50 indoor vs at pasture, respectively, with a low variability across repetitions and no effect of the interval between first and last CSM. If performances of GF measurement indoor or in small plots of intensive pasture are documented, it is the first time at our knowledge that their performances were studied under extensive grazing conditions on natural grasslands in Europe.

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Poster 16

Quebracho condensed tannins fed to dairy goats: effect on milk fatty acids and their use for CH₄ prediction

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The aims of this study were: I) to test the effect of different levels of dietary inclusion (0, 2, 4, 6%) of condensed tannins (CT) extract from quebracho (QE, 71% CT) on goat milk fatty acids (FA), II) develop prediction equations for CH₄ emission of goats using milk FA. A repeated 4×4 Latin square design was applied using 8 goats housed in respiration chambers where CH₄ emission was measured. The diets were composed by a dry basal ration plus a cereal meal supplement containing the different levels of QE. Milk was analysed for FA by gas chromatography. The effect of QE on FA composition was statistically tested with a mixed procedure, considering as fixed effects the experimental square, period, and treatment, with the animal selected as random effect. Multivariate analysis was performed using the stepwise procedure. Only variables with P<0.1 entered the model, and variables with P<0.05 were retained in the final model. The inclusion of QE increased polyunsaturated FA and the ratio cis-9 14:1/(14:0 + cis-9 14:1), considered the best proxy of the desaturation activity of mammary Stearoyl Co-A desaturase enzyme. Multivariate analysis resulted in equations: CH₄ (g/kg DMI) = $37.7 (\pm 2.97) - 2.87 (\pm 1.15) \times C6:0 - 129 (\pm 19.3) \times trans-6,8\ 18:1 + 40.6 (\pm 10.1) \times trans-12\ 18:1 + 11.9 (\pm 4.26) \times cis-11\ 18:1 - 7.76 (\pm 1.96) \times cis-9, cis-12, cis-15\ 18:3 - 85.4 (\pm 27.0) \times cis-5, cis-8, cis-11, cis-14, cis-17\ 20:5$ (adjR² = 0.88), and CH₄ (g/kg FPCM) = $19.3 (\pm 2.38) - 5.21 (\pm 1.21) \times trans-11\ 18:1 + 19.3 (\pm 6.47) \times cis-9\ 10:1 - 9.54 (\pm 3.76) \times cis-11\ 18:1$ (adjR² = 0.83). Milk FA showed a good potential to predict CH₄ emissions in goats.

Dietary incorporation of Zinc-Beta glucan improves immune status, gut microbial composition, and mitigates gas emissions in weaning pigs

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Zinc oxide (ZnO) is currently used as a dietary supplement to support gut homeostasis during the standard 'abrupt' weaning practices in commercial pig production. Besides, glucans are used as broad-spectrum immunopotentiators to improve gastrointestinal (GI) health milieu by increasing mucosal barrier functions and contributing to increased animal growth. However, a mechanism through which glucan supplementation of pig food formula improves pig growth performance is unknown. Thus, this study aimed to explore the potential effects of dietary administration of zinc-beta glucan (ZB) on productivity, nutrient digestibility, gas emissions, blood profile, and microbiome analysis in weaning pigs. A total of 160 weaned piglets (6.38 ± 0.36 kg) were randomly assigned to four treatment groups for 6 weeks. The test treatments were: TRT1, basal diet (control); TRT2, basal diet + 0.25% antibiotic; TRT3, basal diet + 0.05% ZB; TRT4, basal diet + 0.1% ZB. Feeding weaning pigs with 0.1% ZB reduced NH₃ emissions and increased lymphocyte levels in the blood. The addition of ZB improved ($P < 0.05$) alpha and beta diversity in weaning. Furthermore, taxonomic distribution analysis showed highly abundant phyla Firmicutes and Bacteroidetes and reduced ($P < 0.05$) pathogenic bacteria in ZB group pigs. The findings suggest that the inclusion of 0.1% ZB would be beneficial to enhance the immune status and reduce the gas emissions in pigs by altering their gut microbial composition. This study provides valuable insights into ZB supplementation as a novel strategy to improve gut health and performance in pigs through dietary modification.

Enterococcus gallinarum Reduce Methane Production in Rumen Fermentation In Vitro

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CH₄ is a major end product of anaerobic fermentation in the rumen and a greenhouse gas and reduction of CH₄ emissions is desirable. This study was conducted to isolate and identify Enterococcus strains and to determine their reduction of CH₄ emissions activities. Enterococcus strains were isolated from rumen fluid samples from Korean Holstein cow. Enterococcus strains were identified as Enterococcus gallinarum using 16S rRNA gene sequencing and the NCBI BLASTn program. The generated CH₄ concentration was significantly lower in the Enterococcus gallinarum treatment groups after 24 h of incubation in rumen fermentation in vitro ($P < 0.01$). Ruminant contents were obtained from cannulated Holstein cow with body weight of 600 ± 47 kg that was fed twice a day with feed concentrate and rice straw at a 2:8 ratio. The inoculant was prepared by mixing the rumen fluid with a McDougall's buffer medium (pH 6.9) in a 1:2 rumen fluid:buffer ratio. Enterococcus gallinarum strains (1%) was then anaerobically inoculated into serum bottles and incubated in a shaking incubator at 39°C with horizontal shaking at 100 rpm. The in vitro fermentation parameters were monitored for 24 h. Gas samples were analyzed for CH₄ using gas chromatography with TCD-Methanizer-FID. The one-way analysis of variance (ANOVA) was used to analyze statistical differences among multiple groups using Tukey's honestly significant difference (HSD) test as the post-hoc test. These findings indicate that Enterococcus gallinarum has potential as a direct-fed microbial additive for decreasing CH₄ production.

Insights into Effects of Combined Capric and Lauric Acid on Rumen Microbial Composition

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Previously, the combination of low doses of capric acid (C10) and lauric acid (C12) showed the potential to decrease methane (CH₄) emissions from cattle, but also negatively affected nitrogen utilization in the rumen. However, its mode of action on rumen microbiota is not clear. Therefore, the objective of this study was to investigate the combined effects of C10+C12 on rumen microorganisms. Briefly, eight Holstein cows were randomly assigned into two groups in a cross-over design and were fed two diets based on silage with the addition of either 100 g of stearic acid per cow/d (control), or 50 g of C10 combined with 50 g of C12 per cow/d (C10+C12). The composition of microbiota in the rumen fluid was assessed using NGS. The data from NGS was explored through downstream analysis in R and Python, including statistical analysis using Wilcoxon signed-rank test. Alpha diversity indices and the ASV counts were not affected ($P>0.05$) by C10+C12. The relative abundance (RA) of Archaea was not affected ($P>0.05$) as well, which suggests that decrease in methanogenesis could be caused by some other mechanism than the direct effect on RA of Archaea. The most significant effect of C10+C12 was on the RA of Clostridia, which increased ($P<0.05$), and RA of Prevotella, which decreased ($P<0.05$). Prevotella are controversial and diverse microorganisms, in some studies connected with lower CH₄ emissions, and in some research associated with high CH₄ yield. The increased representatives of Clostridia may have increased ammonia concentrations because some members of this class are hyper-ammonia producing bacteria.

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Enhancing Ruminant Fermentation Efficiency: Mitigating Methane Emissions through Persistent Peroxide Addition

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Agricultural methane accounts for approximately 20% of all greenhouse gas emissions. Peroxide compounds have been demonstrated to effectively reduce methane emissions from ruminants. This study explored the effects of persistent vs pulse dosed Hydrogen Peroxide (H₂O₂) and Calcium Peroxide (CaO₂) to reduce CH₄ production and increase animal performance in an in vitro rumen model. Batch rumen incubations in 1-L Schott bottles were prepared with rumen fluid and anaerobic buffer. Batch incubations were continuously fed over a 48-hour duration with a liquid substrate. The effect of persistent peroxide compound addition over time was compared to a pulse dose (once per day), whilst maintaining the same total peroxide concentration over the 48-hours. Rumen fermentation dynamics were monitored for biogas production, methane concentration, oxidation-reduction potential (ORP), volatile fatty acids (VFA's), chemical oxygen demand (COD) and microbial consortium. Methanogenic archaea function best in an ORP environment of $-300 > x < -600$ mV. Persistent peroxide addition had longer lasting elevated ORP levels and enhanced inhibition of methanogenic archaea for longer periods compared to the one-time pulse treatment. When adding small amounts of 3% H₂O₂ frequently throughout the first 12 hours of 48-hour experiments, 58% reductions were seen in treated vs control groups along with a higher VFA accumulation which may be due to hydrogen being driven towards VFAs and not CH₄. Chemical Oxygen Demand (COD) levels were similar in treated vs control suggesting digestibility was unaffected. Overall, H₂O₂ and CaO₂ showed promise in reducing methane and altering fermentation in an in vitro model.

Performance evaluation and enteric methane emission in bovines treated with natural Fator P additive

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Zootechnical additives in bovines diet modulate the fermentation ruminal and is essential to increase the efficiency of productive and sustainable. The use of natural additives, require research that validates the effectiveness in productivity and reduction of enteric methane emissions. The Fator P is natural additive, compound by symbiotic and fatty essential acids, as a substitute of tradition additives. The aim was value the methane enteric emission of animals treated with and without the Fator P, in gases collected through respirometric masks and analyzed by chromatography gas. Were used 24 animals of Nelore breed, ages between 18 and 24 months, average weight of 500kg, conditioned for the experimental routine. After adaptation, the animals received 2,5% of corporal weight in dry matter (DM) in diet compound of 50% of concentrated and 50% of roughage. Two groups were divided (group control and Fator P) based on methane gas (CH₄) evaluated in the pre-initial phase. The results, expressed in grams (g) of CH₄ per kilogram (kg) of DM ingested, of difference between control group (15,271g CH₄ / kg DM) and Fator P (13,286g CH₄ / kg DM) showed a reduced in 1,985g CH₄ / kg DM ingested (p value = 0,0349*). In carbonic gas equivalent (CO₂ eq), the difference can be up to 45,7g / kg DM ingested. Considering the average dairy gain (ADG) of 1,318kg and 1,171kg respectively, the Fator P group and control, the difference of CO₂ eq / kg of ADG was to 851,5g less for Fator P group, showing that this additive improves the productive and sustainable efficiency of livestock farming.

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Utilizing a Feed Additive to Mitigate Methane Emissions from Dairy Cows: On-Farm Trial Employing Methane Laser Detector

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Novel strategies to modulate methane emissions from ruminants, such as ruminal manipulation with feed additives, are crucial for reducing environmental impact of livestock. At the same time, adopting smart technologies for methane measurement allows to evaluate the on-farm efficacy of feed additives. The current study employed the Laser Methane Detector (LMD) to assess the effects of a feed additive composed of tannins and essential oils. Twelve Holstein cows were randomly allotted to control diet (CON) or control diet with feed additive (FAD), in order to obtain 10 mid-lactation cows for group, balanced according to milk yield (20.3±3.2; 21.4±3.2 L), weight (693±68; 689±48 kg), days in milk (145±61; 145±55), and parity (2.0±1.1; 2.0±0.9). Additive administration was top dressed and lasted 10 weeks. Individual methane emission was measured using LMD in week 0 and from weeks 6 to 10, six times per week for five minutes each. Methane data were analyzed with a linear mixed model incorporating treatment, period, and animal as random effect. Generally, methane emissions varied across weeks but remained consistent between treatments. At week 10, no significant differences were observed, 403.1±11.8 and 412.8±13.7 g/d, for CON and FAD respectively. Methane emission per liter of milk also showed no variation, 24.96±1.1 and 26.25±1.7 g/L, for CON and FAD. Further analysis of daily methane emission trends is on-going to explore the impact of supplementation on emissions patterns.

In vitro and in vivo assessment of a blend of chestnut and quebracho tannins on rumen fermentation, methane production and digestibility in ruminants

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The objective of this work was to assess in vitro and in vivo the potential of a blend of chestnut and quebracho tannins (CQT) to modify rumen fermentation, methane (CH₄) production and total tract apparent digestibility. First, a 24-h in vitro batch culture experiment was conducted using rumen fluid from 4 cows to test 4 levels of inclusion of CQT (1, 1.5, 2 and 4 % of diet) to evaluate effects on production of gas, volatile fatty acids (VFA) and CH₄. Second, an in vivo trial was conducted to evaluate the effect on CH₄ production and digestibility using 32 adult sheep that were randomly allocated to one of the following experimental groups (n=8): control and CQT added at 0.225, 0.450 and 0.675 % of diet DM. Animals were adapted to the diets for 14 days, then the production of CH₄ was determined using open-circuit respirometry chambers during 3 consecutive days and total tract digestibility estimated using Manganese as internal marker in feed and faeces. Inclusion of CQT significantly (P<0.001) reduced the production of CH₄ in vitro (6.95, 6.12, 6.02, 5.69 and 5.27 ml CH₄/g DM incubated, respectively, for control and the four levels of CQT) without affecting (P>0.05) total gas production or total VFA concentration. In vivo, the inclusion of the blend did not modify DM intake, total tract digestibility, or CH₄ emissions. To clarify the inconsistency between in vitro and in vivo results, the effect of inclusion level, basal diets characteristics and treatment periods merits further investigation.

Can almond hulls reduce in vitro ruminal methane production?

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Almond hulls (AH) are the main by-product of almond production and are frequently used in dairy ruminant feeding, but information on its effects on enteric CH₄ production is controversial. This study aimed to assess the in vitro ruminal fermentation and CH₄ production of diets for dairy ruminants including increasing amounts of AH. The AH sample included in the diets contained (as dry matter (DM) basis) 7.70, 7.37, 34.7 and 12.1% of ash, crude protein (CP), neutral detergent fiber (NDF), and total sugars, respectively. Four experimental diets having similar CP (16.6%) and NDF (37.0%) content (DM basis) were formulated: a control diet without AH, and 3 diets including 8, 16 and 24 of AH (fresh matter basis). All diets contained 40% alfalfa hay, and the AH partially substituted corn grains, wheat bran, and sugar beet pulp in the control diet. Diets were fermented in vitro with buffered sheep ruminal fluid for 24 h to measure fermentation parameters (pH, volatile fatty acids (VFA), NH₃-N) and CH₄ production. Increasing the AH level reduced linearly (P ≤ 0.026) the production of gas, total VFA and CH₄, the concentrations of NH₃-N and the molar proportion of butyrate, but increased final pH (P < 0.001; linear). These results indicate a reduction of diet fermentability by including AH. In fact, the ratio CH₄ / total VFA was similar for all diets (P = 0.642 and 0.376 for linear and quadratic effects, respectively), confirming that the observed reduction in CH₄ production was due to decreased ruminal fermentability of the diets containing AH. The results indicate that the used AH did not contain antimethanogenic compounds.

Anti-methanogenic feed additives in pasture-based dairy systems

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Additives been widely proven to reduce enteric methane (CH₄) by ~ 30% when mixed throughout a total mixed ration fed to cows in indoor production systems. However, there has been limited research to-date on feed additives in pasture-based systems. In order for additives to have practical application in grazing systems they must be capable of being fed through in-parlour feeders so they can be supplemented twice daily during milking without increasing workload on farms. At present, the most effective feed additives are sensitive to heat and pressure and, as such, unsuited to their incorporation within concentrate pellets during the pelleting process. However, a novel calcium peroxide (CaO₂) based feed additive, which is reported to effectively reduce enteric methane when fed twice daily to beef steers on a grass silage based diet, is capable of withstanding pelleting without detriment to additive efficacy. A study was undertaken to evaluate the methane mitigation potential of CaO₂ when fed to grazing dairy cows in late lactation. Treatment cows were supplemented with CaO₂ twice daily at morning and evening milking, while control cows received no additive supplementation. Methane was measured using Green-Feed units, and milk production, body weight and body condition score were monitored to determine the effect of additive supplementation on productivity. There was no effect of CaO₂ supplementation on CH₄ output, however, there was an interaction between treatment and parity such that the additive was more effective in parity one and parity two animals compared to those in parity three.

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The Effect of Ensiling Alfalfa with Acacia (*Robinia pseudoacacia*) Tree Leaves and Sugar Beet Molasses on Silage Fermentation Quality, In-vitro Ruminal Fermentation and Methane Production*M. U. Hassan¹, M. Gül¹**¹ Atatürk University, Department of Animal Nutrition and Nutritional Diseases, Üniversite, Atatürk Üniversitesi Kampüsü, Yakutiye, 25030 Erzurum, Turkey*

Agroforestry plant leaves notably woody perennials, are widely used to feed animals in many regions of the world, particularly in the tropics. This study also demonstrated a socio-economic and climate viable impact of ensiling alfalfa with agroforestry leaves. Acacia is a fast-growing leguminous tree that produces a huge mass of leaves rich in protein (30.43%) and condensed tannins (12.5%), as a waste which can be utilized as a ruminant feed source. In fact, alfalfa is difficult to ensile because it contains a low level of water-soluble carbohydrates, high buffering capacity, prone to extensive proteolysis and nutrient losses. In this study, supplementation of alfalfa silage (AAS) with acacia leaves and molasses significantly decreased NH₃-N production and butyric acid content while increasing the acetic acid and dry matter contents of the silage. In the Hohenheim technique, 5% molasses (M5) increased the net gas production, improved the in vitro dry matter degradability and true NDF degradability; meanwhile, reduced the organic matter degradability during daisy incubation of alfalfa silage compared to the control. Moreover, the addition of acacia leaves with three different levels significantly lowered CH₄ production, total VFAs, and ruminal NH₃-N (an indicator for N excretion & environmental pollution) concentrations in a dose-dependent way. The results illustrated that lower level of acacia leaves (3%) in combination with molasses (5%) applied prior to ensiling alfalfa were more useful in reducing silage pH and ruminal NH₃-N ($p < 0.001$), improving the relative feed value and quality ($p < 0.001$), metabolizable and net energy lactation of AAS ($p < 0.001$) without any adverse effect on degradability parameters compared to control.

Temporal effects of botanicals on total gas and methane production in ruminal fermentation in vitro
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The purpose of this study was to evaluate the temporal effect of botanical additives on gas production in the automated Gas Endeavour® in vitro system of ruminal fermentations. Three different natural identical compounds were selected: anethol (ANT), carvacrol (CAR), and thymol (THY) were tested in triplicate at 100 ppm and with 8 replicates at 250 ppm. A control group was included in both studies. Rumen fluid, obtained from non-lactating Holstein cows, was used as inoculum for a 24h in vitro fermentation. Measurements of total gas and methane production were taken every hour. Accumulated 24h data were analyzed using a two-way ANOVA and hourly data as a repeated measurement with multiple comparison as a post-hoc analysis. At 100 ppm CAR, THY and ANT tended to reduce methane production ($P < 0.15$). At 250 ppm, total gas was reduced starting at h 5 (Treatment x time, $P < 0.05$) with -66,63%, -63,88%, and -70,69% reduction (respectively for ANT, THY, and CAR) at h 24 compared to control. Similarly, methane reduction started at h 3 (Treatment x time, $P < 0.05$) with -50,25%, -32,03%, and -71,43% reduction (respectively for ANT, THY, and CAR) at h 24 compared to control. We conclude that, at 250 ppm concentration, ANT, THY and CAR were able to significantly reduce total gas and methane production within the first hours of in vitro fermentation, in particular carvacrol showed the greatest effect.

High throughput in vitro screening of natural essential oils, rumen buffers, and plant extracts for enteric methane mitigation in cattle

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Ruminant dietary additives were investigated for enteric methane (CH₄) mitigation and their effects on rumen fermentation characteristics post in vitro supplementation. Essential oils, natural compounds, and rumen buffers were compared at optimum dosages with commercial products via high-throughput screening as a follow-on study from a meta-analysis review of potential feed additives that employ diverse enteric methane-mitigating approaches. Investigated feed additives included *Allium sativum* extracts garlic oil (GO), and garlic powder (GP), *Asparagopsis* (ASP), calcareous marine algae (CMA), calcareous marine algae fortified with magnesium oxide (CMA.MgO), essential oils (EO), and *Yucca schidigera* plant extract (Yucca). Screening of potential additives at optimum dosage revealed promising effects on rumen parameters, total gas, CH₄ levels, and volatile fatty acid (VFA) profiles. Rapid high-throughput screening of feed additives reveals the greatest potential for rumen buffers and their combination with essential oil at their lowest optimum dosages. There were significant decreases among treatments CH₄ levels when compared to an average of controls with potential for on-farm mitigation with the CMA.MgO rumen buffer, and *Allium sativum* derivatives GO and GP in combination with the CMA rumen buffer. Instances of increased total VFA concentrations with Yucca, and rumen buffers CMA and CMA.MgO reveal their potential to increase ruminants' primary energy source while also mitigating the energy loss linked to enteric CH₄ production. Ruminant feed additives CMA.MgO, GO, and Yucca display potential for improved rumen productivity based on VFA profiles, and greater CH₄ mitigation when compared to a commercially available blend of EOs designed for mitigating enteric CH₄, following 24 hours of in vitro rumen simulation. Maintenance of rumen fermentation parameters while inducing mitigation of harmful greenhouse gas CH₄ reveals promise for calcareous marine algae sourced rumen buffers as CH₄ mitigators.

Effect of using essential oils blend on performance in dairy cows

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The study investigated the effects of supplementing lactating dairy cows with Agolin Ruminant®, an essential oil blend, on dairy cow performance. A total of 187 Holstein dairy cows were allocated to two dietary treatments based on parity, days in milk (DIM), and milk yield (MY). The control group (n=94; parity 2.14, DIM 121, MY 36.55kg) received basal total mixed ration (TMR), while the Agolin group (n=93; parity 2.16, DIM 120, MY 34.74kg) received basal TMR supplemented with Agolin Ruminant® at a dosage of 1g per cow per day. The trial lasted 12 weeks, including a 3-week adaptation period and a 9-week experimental phase (from July to October, 2023). Measurements included daily group milk yield and composition, as well as weekly manure sieving on a subset of animals from each group. Additionally, a subgroup of 14 cows per treatment group was selected for individual measurements of weekly milk yield and composition (fat, protein, lactose, urea, and somatic cell count), ensuring parity, DIM, and milk yield balance. Average MY was 33.63Kg in the control group vs 34.76Kg in the treated group (+1.13Kg, p<0.001). Average Fat Protein Corrected Milk (FPCM) was 33.6Kg in the control vs 34.41Kg in the treated group (+0.81Kg, p<0.01). Average fat yield was 1.33Kg in the control vs 1.37Kg in the treated group (p<0.001). The results in the subgroup analysis are as follows: +1.71Kg of milk in the treated group, p<0.1 (34.01Kg in the control vs 35.72Kg in the treated group). +2.79Kg of FPCM in the treated group, p<0.01 (33.86Kg control vs 36.65Kg treated). +6.8% fat% in the treated group, p<0.1 (3.98% in the control vs 4.25% in the treated group). Furthermore, improved manure scores suggested enhanced diet digestion in the treated group. The farm's emissions have been calculated using a comprehensive model, Alltech E-CO₂, which suggests that Agolin supplementation could decrease emissions intensity by 6% to 12% compared to the control scenario. This reduction is primarily attributed to an increase in FPCM. In conclusion, Agolin Ruminant® supplementation appears to be a promising strategy for simultaneously enhancing dairy production efficiency and mitigating methane emissions.

Culture and growing conditions has an impact on methane mitigation properties in red algae

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The study, conducted in partnership with a Swedish algae cultivation company, aimed to assess the methane inhibiting potential of *Asparagopsis* spp. (AS) under various cultivation conditions. Six land-based algae cultures and two wild-harvested batches were tested in vitro using a common dairy cow diet composed of 60:40 silage:concentrate on DM. The algae, constituting 0.5% of dietary OM, were incubated with rumen fluid in serum bottles. All treatments, including a control diet (CON) and blank bottles, were conducted in triplicate and repeated in two runs. An automated in-vitro gas production system continuously recorded gas levels every 12 minutes throughout the incubation period of 48 hours. Gas samples were injected into a star 3400 CX series gas chromatograph (Varian Chromatography, USA), at five different time points; 2, 4, 8, 24 and 48 hours of incubation and were used in calculations to predict in vivo methane production. Statistical analysis, accounting for the random bottle effect and incorporating fixed effects for run and treatment, was used to assess significance. The predicted in vivo methane production from the 48-hour gas analysis of the CON was determined to be 48.1 (± 4.0) mL/g OM. Significant treatment effects (P<0.001) were observed in four of the cultured algae, reducing methane production by 21%, 23%, 25%, and 83% respectively. The other tested algae did not show any significant impact on methane production. This research contributes to enhance knowledge about red algae strategies for sustainable methane reduction in livestock by showing that different strategies of cultivation results in different methane inhibiting capacity.

Freshly cut grass-clover vs. grass-clover silage and grass-clover pulp silage for organic growing-finishing pigs
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This study assessed the impact of feeding organic growing-finishing pigs with freshly cut grass-clover versus grass-clover silage and grass-clover pulp silage from 30-115 kg. The experiment also measured the impact of reducing the protein content in concentrates by 10%. The study involved 270 pigs, and production parameters, such as daily gain, feed consumption, feed efficiency, meat percentage, blood parameters, meat quality, and eating quality, were assessed. Design: The experiment was performed at the Organic Platform at AU-Viborg, with 270 DLY or LYD pigs of DanBred-origin. The rearing period from 30-115 kg lasted 13-14 weeks. Each pig received 200 Scandinavian Feed Units (FU) of concentrate and approximately 20 FU of roughage. Key results include: Pigs from 85-115 kg had a daily intake of 870 g/d of fresh grass-clover, corresponding to 5% of the daily SID lysine requirement and 13% of the daily CP requirement. Grass-clover silage resulted in a daily intake of 335 g from 85-115 kg., which corresponds to 1.15% of the daily SID lysine requirement and 4.7% of the daily CP requirement from 85-115 kg. No differences were observed in acidity, harshness, meat flavour, metallic taste, bitterness, juiciness, chew resistance, chewing time or pork tenderness between roughages or protein levels. Grass pulp silage can be utilized as well as normal grass-clover silage, and pigs willingly consume it. There is a large individual variation in the consumption of roughage between organic pigs. During summer, the SID lysine content in concentrate can be reduced by 10% from 7.0 g/FU to 6.3 g/FU with simultaneous provision of fresh grass without negatively affecting productivity.

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Theatre 1

Variation of zinc release from phytate by phytase is dependent on the level of dietary Zn supplementation in pig: a meta-analysis

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Phytic phosphorus is known as an antinutritional factor impairing microminerals absorption in pig, such as zinc (Zn) leading to Zn output in the environment. Zinc homeostasis is maintained with the regulation of Zn absorption and endogenous intestinal excretion. Previous studies have shown the positive effect of phytase on Zn digestibility in low Zn diet. This study quantifies the interaction between dietary Zn level and exogenous phytase on apparent total tract digestibility (ATTD) of Zn, and plasma Zn concentration using a meta-analytic approach. A total of 30 studies conducted between 1994 and 2021 were included. The meta-analysis was conducted using a mixed effect model in R studio. The meta-design illustrated that increasing adaptation period to the diet decreased the ATTD of Zn ($P < 0.001$). Therefore, observations were weighted by duration of adaptation period to the diet. The study was limited to data under 250 mg/kg of Zn. Phytase has positive effect on ATTD of Zn ($P = 0.001$; $RMSE = 2.56$) but the interaction between phytase and study effect shown a large variability of the response ($P < 0.001$; CV (%): 167 % of mean slope). However, behind variability, a negative interaction between Zn and phytase ($P = 0.04$) exist. Inclusion of 500 FTU/kg of phytase in two diets containing 40 mg/kg or 120 mg/kg de Zn increased the ATTD of Zn of 15.6% and 5.5% respectively. The same interaction was found between dietary Zn and phytase on plasma Zn concentration ($P = 0.02$). This result reflect the well-known zinc homeostasis mechanisms that allow the pig to adapt its utilization based on the dietary Zn levels.

The combination of an alternative zinc source and low ABC-4 diets can replace the high levels of ZnO in nursery piglet diets

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High zinc levels in piglet diets promote growth and reduce post-weaning issues, yet the high ABC-4 value of Zn (21,863 meq/kg) can increase stomach pH and impair nutrient utilization. Recent formulations have reduced ABC-4 values (200 to 300 meq/kg) as an alternative to high ZnO diets, without impairing piglet performance. This study evaluated a novel zinc source, HiZox® (HZ), in low ABC-4 diets. Using 240 male piglets (DNA 200 × 400, BW 5.90 ± 0.05 kg) in a two-phase feeding regimen, the treatments were: Negative Control (NC, 150 ppm Zn HZ), Positive Control (PC, 3,000 ppm ZnO (phase 1) and 2,000 ppm ZnO (phase 2), Low HiZox with 500 ppm (phase 1) and 300 ppm (phase 2) Zn HZ, and High HiZox with 800 ppm (phase 1) and 500 ppm (phase 2) Zn HZ. Growth performance and fecal scores were assessed using GLIMMIX procedure of SAS, revealing that, in phase 1, High HiZox improved ADG and BW at d10. Moreover, increasing HZ levels quadratically improved ADG, ADFI, and F/G. During phase 2, although no significant differences were observed, there was a tendency for a quadratic increase in ADFI with increasing HZ levels. Overall, there was a tendency for higher ADG with High HiZox, while increased HZ levels quadratically improved ADG and ADFI. There were no significant differences on fecal DM and consistency among treatments. In conclusion, the use of HZ as an alternative to high ZnO maintains comparable performance and fecal consistency while enhancing growth performance, making it an effective replacement for high dietary ZnO in nursery piglet diets when combined with the lower ABC-4 concept.

Session 35

Theatre 3

Nursery pigs kept in poor sanitary housing conditions benefit from diets with low buffer capacity

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Protein digestion and pathogen survival in digesta are controlled by a low stomach pH which is hypothesized to reduce post-weaning diarrhea. This is especially important in nursery pigs because they lack the capacity to secrete sufficient hydrochloric acid in their stomach and even more important in poor sanitary housing conditions (PSC). We aimed to determine the dose-response relation between dietary buffer capacity (BC) and growth performance of nursery pigs kept in PSC. A total of 4 treatments were included: (1) BC290, (2) BC370, (3) BC450, and (4) BC530 mEq/kg. Wheat-barley based diets were fed during d0 (weaning) to d14 followed by one common diet (BC of 550 mEq/kg) until d38. Differences in BC were established by changing calcium sources and organic acids. Each diet was offered to 20 pens with 3 pigs (23 ± 1 days old and 6.8 ± 1.3 kg at weaning) per pen. The PSC were established by lowering environmental temperature by 2°C relative to the standard curve and spreading ~1.5 kg manure/pen from a previous batch of nursery pigs on d0, 4, 7 and 12. Daily body weight gain (DG) and feed intake (FI) were determined on d14 and 38 and feed efficiency (DG:FI) was calculated. Data were analyzed using mixed models using polynomial orthogonal contrasts to examine linear and quadratic responses. Results showed a negative linear effect for FI ($P_{\text{linear}}=0.05$) and DG ($P_{\text{linear}}=0.09$) d0-14 without affecting feed efficiency ($P_{\text{linear}}>0.05$). The effect was caused by a greater DG and FI for BC290 compared to the other diets (183 vs 160 g/d and 206 vs 184 g/d, respectively). There was a carry-over effect into the second phase resulting in a 3% higher BW ($P_{\text{model}}=0.16$) on d38 for pigs fed BC290. Thus, pigs kept in PSC appear to benefit from a diet with low BC.

Effect of zinc on phosphorus and calcium digestibility in post-weaning pigs: a meta-analysis

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Phosphorus (P) is essential to support growth and bone mineralization but has to be supplied close to requirements to minimize P output in the environment. In pig, microbial phytase is commonly used to degrade phytic phosphorus (PP) but in-vitro data indicated that Zn may inhibit P release from PP while in-vivo data indicated pharmacological zinc (Zn) to decrease P apparent total tract digestibility (ATTD). The aim of this study was to quantify the effects of dietary Zn concentration on P and calcium (Ca) ATTD in post-weaning. From a systematic literature search (March – July 2023), a meta-analysis was carried out on 16 publications reporting 47 treatments that have measured P and Ca ATTD. The meta-analysis was conducted using a mixed effect of the experiment in R. The X variables tested were plant and microbial phytase, dietary Zn, and PP. Plant phytase had no effect. Increasing the dietary Zn decreased the P ATTD ($P < 0.001$; RMSE=0.28), the concentration of P in the plasma ($P = 0.01$; RMSE=3.46), and Ca ATTD ($P < 0.001$; RMSE=0.26). As expected, there is a positive effect of microbial phytase on P ATTD ($P < 0.001$) and Ca ATTD ($P < 0.001$), but not for plasma P. Also, interaction was found between Zn and phytase ($P = 0.001$). The current results showed that high Zn level reduced P and Ca ATTD with impacting the efficiency of phytase. For Ca ATTD, the reduced absorption with high Zn supply may be also due to competition for channels in the enterocytes of the small intestine. However, for P ATTD the mechanism remains unclear and need further research, but it is possible that Zn binds to phytic phosphorus and also prevents the action of intestinal phytase.

Effect of full matrix application of a novel phytase without or with a multi-enzyme complex containing xylanase, β -glucanase, amylase and protease on performance of piglets

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An experiment was conducted to evaluate the application of energy and nutrient matrix of a novel consensus phytase variant (PhyG) in combination with a multi-enzyme complex (MEC) on performance of piglets after weaning. Growth performance (ADG), feed intake (ADFI), feed efficiency (FCR) and cost savings were used as response criteria. In total, 234 piglets with 8.20 kg body weight (BW) were used. They were distributed into 13 complete blocks of sex and BW, and allocated in 78 pens for a 34-d trial. Intra-block pigs were randomly distributed to 3 treatments corresponding to control diets (0.40 and 0.33% digestible P in prestarter and starter phases, respectively), a PhyG treatment with diets supplemented with 1500 FTU/kg of PhyG and reduction of dig P, Ca, dig AA and net energy based on the phytase contribution, or a third PhyG+MEC treatment supplemented with the same PhyG dose and MEC (providing 4000 U/kg Xylanase, 200 U/kg β -glucanase, 100 U/kg α -Amylase, and 2000 U/kg Protease), with corresponding matrix value for both phytase and MEC. Diets were based on maize, soybean meal and rice by-products, and wheat middlings and soybean hulls were used to adjust major nutrient content of feed formulas. Production cost savings were determined applying EU costs of raw materials. Overall, an ADFI, ADG and FCR of 703 g/d, 502 g/d and 1.402 were observed, respectively. The enzyme treatments with matrix application showed similar feed intake, growth performance or feed efficiency during the prestarter and starter phases ($P > 0.1$). A reduction of 4.0 and 6.6% reduction on feed cost per piglet raised, or 5.7 and 7.2% reduction per kg of BW produced was determined due to PhyG or PhyG+MEC supplementation, respectively. It can be concluded that application of the nutrient full matrix concept of a novel PhyG phytase and MEC allows to maintain performance and resulted in cost savings on postweaning piglet production.

Kinetics of bone mineralization and allometric changes in response to calcium and phosphorus deficiency in broiler chickens

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Data on growth performance and bone mineralization response to different dietary supplies of calcium (Ca) and phosphorus (P) in broilers is important for developing recommendations that avoid P overfeeding to minimize P excretion. The aim of this study was to evaluate the effect of different levels of Ca and non-phytate P (nPP) on the growth performance and kinetics of soft tissues and bone deposition into the skeleton of broilers. A total of 360 Ross 308 broilers were randomly divided into each of 10 replicate cages of the three dietary treatments. During each experimental phase (0 to 9d, 10 to 21d and 22 to 35d), broilers were fed a control diet (C), a moderate (M) or a low Ca and nPP diet (L), formulated to contain, respectively, 100%, 85% and 75% of the estimated of Ca and of nPP requirements to maximize bone mineralization coming from a recent mechanistic model. Bone mineral content (BMC) was measured in one bird per cage (always the same) anesthetized at the end of each feeding phase by dual-energy X-ray absorptiometry. Allometric regression analysis was performed to predict BMC in relation to BW. Growth performance and lean mass in the L vs H diet was not different at d-9, tended to be lower at d-21 ($P < 0.09$) and were lower at d-35 ($P < 0.01$). Birds fed both deficient diets (L and M) had reduced BMC (respectively, -20% and -14%; $P=0.012$) vs birds fed H diet at d-35. Daily BMC deposition did not differ among L and M diets, while it tended to be higher for H diet (9 to 21d, $P=0.098$; 22 to 35d, $P=0.079$). Similarly, the slope of the allometric regression of the whole-body BMC in the H diet was 16 and 18% higher than in L and M diets ($P < 0.01$), respectively. These findings suggest that modifying dietary Ca and nPP while keeping similar ratio affects growth performance and bone mineralization in the same extent which will important to consider for requirements assessment.

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Theatre 7

Assessment of new phytase products through an in-vitro digestion model alongside a broiler chicken trial

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Phytases are widely used enzymes in animal nutrition for their ability to break down phytic acid, the main storage form of phosphorus (P) in plants, making it available to monogastric animals. A new generation of phytase products is offered by suppliers, necessitating comparative evaluation against older or competitor products for practical application. This study aimed at assessing five phytases (4 new products) in a broiler trial and through an in-vitro model. Ten-day old Ross 308 male broiler chickens were randomly assigned to 3 control groups with increasing available P (aP) levels: 0.14, 0.29 and 0.43 (NC, NC+, PC) and 5 groups (A to E) with different phytases added to the NC, each tested at 1.5 times the recommended dose. On day 28, 12 birds per group were euthanized for bone analysis. Additionally, phytases were evaluated using a three-step in-vitro digestion simulation model at 5 doses (0, 250, 500, 750 and 1000 U/kg). Dose response curves were modeled using the equation $Y=a(1-be^{-kx})$. Increasing aP level linearly increased bone ash and P content ($P < 0.001$). Group C showed significantly higher bone P content compared to NC ($P < 0.05$). Bone ash was significantly increased compared to NC in groups A, C and D ($P < 0.05$) but not significantly in group B nor E. Enzymes increased bone P content by 3.35 to 10.05 % and ash by 5.56 to 8.88 % compared to NC with a slightly different hierarchy between products. The in-vitro results showed good correlation ($R^2=0.67$) with in-vivo bone P content. The in-vitro model is a valuable initial assessment tool for new phytase products, necessitating additional in-vivo evaluation, particularly for extra-phosphoric effect on growth performance.

In vitro prediction of calcium and phosphorus availability in monogastric feed using marine sourced calcium and limestone

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Phytate is the principal storage form of phosphorus (P) in feed material and has been shown to reduce mineral availability due to the chelating effect of its phosphate groups. Optimal utilization of nutrients and the reduction of interactions that reduce digestibility by monogastric livestock can provide significant improvements in production efficiency. The aim of this study was to predict calcium (Ca) and phytate P availability in vitro with different Ca sources. A series of digestion simulations were performed using different Ca sources and levels, different commercial phytases and varying inclusion rates with phytate as the only P source. In vitro results demonstrated that 78%, 5%, 9%, and 15% (w/w) phytate P was available in simulations with coarse limestone (D50 ;166µm), fine limestone (D50; 9.22µm), feed grade limestone (D50; 26.3 µm) and a marine mineral complex (MMC) rich in calcium (D50; 33.4 µm), respectively using technical grade phytase. Use of 2 different commercial phytases and varying inclusion rates (3 rates) affected available P ($P<0.05$). A 30% increase in available P was recorded in a diet simulation containing a blend of feed grade limestone (0.15%), MMC (0.4%), and monocalcium phosphate (MCP) (0.4%) compared to a control diet of feed grade limestone (0.55%) and MCP (0.4%) only. Both diets contained an equal phytate amount. Differences in the amount of precipitated Ca-phytate was visible. Overall, the in vitro data demonstrated an inverse relation between available Ca and P. The MMC Ca source reduced interactions between the nutrient sources allowing the phytase to catalyse the release of P more efficiently.

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Theatre 9

Impacts of Saccharomyces yeast postbiotics on intestinal response and growth of nursery pigs

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The small intestine is the main site for the digestion of feeds and absorption of nutrients, but also responsible for immune protection from the external stressors. Upon weaning, feed intake can cause mucosal inflammation, and oxidative damages resulting in mucosal damages and repairing in the jejunum. This study aimed to determine the efficacy of postbiotics derived from Saccharomyces yeast on the intestinal health of newly weaned pigs. Thirty-two newly weaned pigs (16 barrows and 16 gilts) at 21 days of age (6.05 ± 0.24 kg body weight) were assigned into 2 dietary treatments based on a randomized complete block design, with initial body weight and sex serving as blocks. The dietary treatments were a control and a group supplemented with Saccharomyces yeast postbiotics (SYP, 175 g/ton feed). The pigs were fed for 35 days. At the end of each phase, the pigs and feed disappearance were individually weighed to determine growth performance parameters. Fecal score was recorded from day 3 of the experiment. After 35 days of feeding, the pigs were euthanized, and intestinal mucosa was collected from the jejunum to assess immune status and oxidative stress. Jejunal tissue was also collected to measure the expression of genes associated with intestinal barrier markers, cell proliferation and apoptosis. Data were analyzed using SAS 9.4. The SYP did not influence the growth performance of nursery pigs. However, SYP reduced ($P<0.05$) the fecal score from 4-7. The SYP did not affect the production of inflammatory cytokines and oxidative damage products whereas it tended to increase the gene expression of IFN- γ ($P=0.071$) and mTOR ($P=0.080$), decrease the gene expression of BAX1 ($P<0.05$), tended to decrease the gene expression of SGK1 ($P=0.066$), increased ($P<0.05$) cell proliferation in the crypts (Ki-67 positive cells), and tended to increase the villus height ($P=0.078$) and crypt depth ($P=0.052$) in the jejunum. Saccharomyces yeast postbiotics reduced diarrhea of newly weaned pigs and provided protection to the jejunal mucosa by promoting cell proliferation, and reducing the expression of genes associated with apoptosis.

Effect of hydrolyzed yeast from *Kluyveromyces fragilis* replacing high-dose zinc oxide on growth performance, diarrhea incidence and intestinal health of weaned piglets

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High-dose zinc oxide (ZnO) has been using as a substitute for in-feed antibiotics due to its positive effects on the development and intestinal health of weaned piglets, while it would cause serious environmental pollution and limitation of mineral absorption. Hydrolyzed yeast from *Kluyveromyces fragilis* (HK) is rich in a variety of nutrients without toxic and harmful compounds, which have received increasing attention due to their superior physiological properties. In our study, 2 experiments were conducted to investigate the effect of optimal dose of HK as a novel alternative to high-dose ZnO on growth performance, diarrhea incidence and intestinal health of weaned piglets. In experiment 1, one hundred weaned piglets were randomly allotted to 4 groups with 5 replicates in a 21-day experiment. Piglets were fed with basal diet (NC), NC + 2g/kg zinc oxide (PC), NC + 7.5g/kg HK (HK1), or NC + 10g/kg HK (HK2). Dietary HK at 10 g/kg alleviated diarrhea incidence of piglets that might be attribute to the improved plasma antioxidant efficiency and immune status and the regulated fecal microbial community, and could be alternative to the high-dose ZnO. In experiment 2, a total of 160 weaned barrows were randomly allotted into 4 treatments, with a 2×2 factorial design that compared ZnO [sham (-) or ZnO (+)] inclusion under different dietary treatments [basal diet (NC) or NC + 10 kg/t hydrolyzed yeast (HK)]. HK supplementation improved the growth performance and reduced the diarrhea incidence of weaned piglets, which may be attributed to the increased antioxidant and immune levels, reduced intestinal inflammatory response, enhanced intestinal barrier function, improved intestinal tissue morphology, and regulated intestinal microbiota.

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Theatre 11

In vitro and in vivo evaluation of L-Lysine solid lipid microcapsules for swine

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The aim of the study was to evaluate gastric bypass, intestinal slow release, and bioavailability of L-Lysine encapsulated in solid lipid microcapsules (L-SLMs), compared to free L-Lysine, in swine. The pharmacokinetics profile of L-SLMs was characterized in vitro and validated in vivo. In vitro, the release of L-Lys from SLMs was evaluated incubating a 0.5 g sample at 40°C first in gastric phase (pepsin buffer, pH=1.5; 2h) and then in intestinal phase (pancreatin buffer; pH=7.5; 8h). L-Lys quantification was performed via Kjeldahl method. In vivo, 12 weaner piglets (BW=15 ± 2 kg) were fed corn meal and divided in 3 groups (n=4) receiving an oral dose of either saline solution (placebo), free L-Lys at 0.17 g/kg BW (control) or L-SLMs at 0.38 g/kg BW (treatment), which provided the same amount of L-Lys. Plasma samples were collected before oral administration and hourly until 24h. L-Lys was quantified by LC-MS/MS. Data were analyzed using Two-Way ANOVA, with differences considered significant at P<0.05. In vitro, the gastroprotection of L-Lys from SLMs was 84% at 2h, while the intestinal release was 80% at 8h. In vivo, free L-Lys showed a plasma peak concentration of 1054.10 µM after 1h and completely disappeared after 8h. Conversely, L-SLMs displayed a plasma peak concentration of 1184 µM at 3–4h, and then the hematic L-Lys concentration gradually decreased to 0 until 24h. The bioavailability was higher for L-SLMs compared to free L-Lys (AUC=7902 µM×h and 3175 µM×h, respectively). Overall, the administration of L-Lys in SLMs has the potential to guarantee intestinal slow release and enhanced bioavailability of L-Lys compared to not protected L-Lys.

Effect of a botanical-based feed additive in vitro on cultured enterocytes and in vivo on weaned piglets challenged with F4+ enterotoxigenic *Escherichia coli*

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The aim of the study was to evaluate a botanical-based feed additive (BOT) both in vitro on cultured enterocytes and in vivo in weaner piglets orally challenged with F4+ enterotoxigenic *E. coli* (ETEC). In vitro, Caco-2 cells were treated with BOT and infected with 5×10^7 CFU of ETEC (BOT+); enterocytes integrity, and ETEC adhesion and translocation were measured. Unchallenged (CTR-), challenged (CTR+), and antibiotic (colistin, COL+) controls were included. In vivo, 30 F4-susceptible weaner pigs were divided in 3 groups (n=10): positive control (CTR+, no treatments); control with colistin in water (COL+); treatment receiving BOT, microencapsulated in lipid matrix, at 2 kg/MT (BOT+). On d9, each pig was orally challenged with 3×10^9 CFU of ETEC. Body weight (BW), fecal score, and ETEC shedding were recorded until d23. Data were analyzed with ANOVA (in vitro) or ANCOVA (in vivo, covariate: d0 BW), with differences significant at $P < 0.05$ and tendencies at $P \leq 0.1$. In vitro, integrity of CTR+ enterocytes was significantly reduced (26% of starting value), while BOT+ kept it at around 80%, in line with CTR- and COL+. Moreover, BOT+ reduced ETEC adhesion and translocation across enterocytes by $1 \log_{10}$ (CFU/mL) ($p < 0.05$). In vivo, despite BOT+ led to higher overall fecal ETEC shedding vs CTR+ and COL+ ($P < 0.001$), no differences were observed in fecal scores, and BOT+ tended to improve BW compared to both CTR+ and COL+ at d14 (+1.1 kg) and d23 (+1.5 kg). BOT protected enterocytes during ETEC infection in vitro and may support growth of ETEC-challenged piglets.

Session 35

Poster 13

Evaluation of dietary selenium nanoparticles and curcumin nanospheres on growth, feed utilization and fecal gas emissions in weaned piglets

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We investigated the dietary supplementation of selenium nanoparticle (SeNP) and curcumin nanosphere (CN) based on growth, feed utilization and fecal gas emissions in weaned piglets. A total of 108 weaning pigs (Duroc × [Yorkshire × Landrace]) with an average initial body weight of 6.50 ± 1.03 kg were randomized into 9 dietary groups in triplicate pens (4 pigs in each pen). Dietary treatment groups were as follows: 1) CON, basal diet; 2) T1, basal diet + SeNP (0.3 mg/kg); 3) T2, basal diet + SeNP (0.5mg/kg); 4) T3, basal diet + CN (0.5 ml/kg); 5) T4, basal diet + CN (1.0 ml/kg); 6) T5, basal diet + SeNP (0.3 mg/kg) + CN (0.5ml/kg); 7) T6, basal diet + SeNP (0.5 mg/kg) + CN (0.5ml/kg); 8) T7, basal diet + SeNP (0.3 mg/kg) + CN (1.0 ml/kg); 9) T8, basal diet + SeNP (0.5 mg/kg) + CN (1.0 ml/kg). All data were processed by Duncan's multiple range test using the General Linear Model procedure of SAS (2013) to test the significance between the means ($P < 0.05$). At the end of 21-day feeding trial, T8 treatment group was significantly higher in body weight than CON treatment group ($P < 0.05$). During the entire experiment period, T7 and T8 groups were significantly higher in average daily gain than CON group ($P < 0.05$). There was no significant difference in dry matter, nitrogen and energy utilization during the entire study period. At the end of experiment (21 day) pig feces in CON group was significantly higher in ammonia (NH₃) than T3, T5 and T6 treatment group ($P < 0.05$). Overall, the results demonstrated the potential of dietary supplementation of CN and SeNP as a nanobiotechnology tool as well as effective feed additive in terms of improving performance and reducing fecal ammonia emissions in weaned piglets.

The use of levan in laying hens nutrition - influence on the microflora of the gastrointestinal tract

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Levan is an exopolysaccharide which, thanks to its prebiotic properties, can potentially be used as a feed additive in poultry nutrition. The aim of the study was to determine the effect of the addition of various levels of levan on the development of the microflora of the gastrointestinal tract of laying hens. The experiment was conducted on 120 Lohmann Brown laying hens. The birds were divided into 5 experimental groups (8 replications (cages), 24 hens in each group). Group C (control) did not receive any levan supplement. The L0.1 group received 0.1% levan in the feed mixture, the L0.25 group – 0.25% levan in the feed mixture, the L0.5 group – 0.5% levan in the feed mixture, and the L1 group – 1% levan in the feed mixture. The experiment lasted 90 days. At the end of the experiment, 8 hens from each group were euthanized and the contents of the jejunum and cecum were collected for microbiological analyses. Analyses were made of the number of bacteria of the genera *Lactobacillus*, *Bifidobacterium*, *Clostridium* and those belonging to the *Enterobacteriaceae* family. The analysis of the obtained results showed a lower number of bacteria of the *Bifidobacterium* genus in the cecum of birds from the C group. In turn, in the case of *Clostridium* bacteria, a reduction in the number of birds with these bacteria found in their intestinal contents was demonstrated in all groups that received levan added to the feed compared to the C group. The content of the jejunum showed a reduction in the number of bacteria from the *Enterobacteriaceae* family in birds from groups that received levan supplement in the feed. Therefore, it can be concluded that the use of levan in the laying hens nutrition has a positive effect on the development of probiotic microflora while inhibiting the development of pathogens.

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Poster 15

Mint oils: ability to perform in vitro anti-inflammatory and their antimicrobial effect

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Mint oils (MO) due to bio-active molecules with anti-inflammatory and antibacterial properties are being explored to counteract with antibiotic overuse in swine. The aims were to test peppermint (PMO) and spearmint (SMO) oils via (i) measuring in vitro anti-inflammatory effects with macrophage (PAM) cells, (ii) testing the antimicrobial activity against swine pathogen, *Escherichia coli* ETEC F18+ (EC). Briefly, PAMs were harvested from the bronchial lavage and seeded (10⁶ cells/mL). After 24 h (37°C, 5% CO₂), cells were treated with MOs or lipopolysaccharide (LPS) with 2 × 5; 2 doses of LPS (0 or 1 µg/mL) and 5 doses of MOs (0, 25, 50, 100, 200 µg/mL). The supernatants were collected to measure Interleukin-1 beta (IL-1β). Besides, a culture-based EC assay was performed to evaluate the inhibitory activity of PMO and SMO in doses (0, 1.44, 2.87, 5.75, 11.50, 23.0 mg/mL). Overnight-grown EC were treated and measurement (OD₆₀₀, 60 min interval), data were converted (CFU/mL) and analyzed (SAS 9.4). Treatment with PMO and SMO significantly inhibited (p<0.05) IL-1β secretion from LPS-challenged PAMs in a dose dependent manner. Also, EC growth inhibitory results revealed that PMO and SMO inhibited (p<0.05) the growth of EC at different concentrations. Results showed that PMO and SMO had anti-inflammatory activities in vitro and antimicrobial activities against EC, which indicates MO may be candidates to decrease antibiotic overuse.

Update on the use of slow-growing chicken genotypes for conventional and alternative farming systems in the EU

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There is a growing interest in the use of slow growing (SG) and dual-purpose genotypes in alternative (organic, free range) and conventional farming systems in the EU. The reason for this positive trend originates from the increasing attention of public opinion and regulatory agencies toward animal welfare and qualitative traits of production. Although it is crucial to harmonise criteria and procedures in EU, these genotypes are characterised in different EU countries and bodies (e.g. European Chicken Commitment) with different approaches. Many of these approaches consider only the performance of chickens (e.g. daily weight gain), even if the assessment of functional traits in challenging conditions, such as adaption to environmental stress, locomotion activity, animal welfare and health, and immune response should be analysed in detail. Comprehensive information on this topic are recent and several discrepancies in both literature and commercial circumstances render the characteristics of production and the result obtained very different along EU. Although SG, in conventional and alternative systems, are widely used, a whole assessment is lacking. Indeed, the comparison of genetic strains in various farming systems which implies deep changes in input, output and side effects, should be done using multicriteria tools, able to simultaneously measure the effect of a given alternative on a panel of traits. The lecture will analyse the current knowledge on the relation between the main traits implicated in the chicken adaptation to a farming system developing a “one welfare” approach capable to compare animal welfare, quality, sustainability and environmental impact at the same time.

Session 36

Theatre 2

Crossbreeding as an innovative way to preserve local chicken breeds through use in organic farming

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In organic poultry production, dual-purpose chickens are favored to avoid the killing of male chicks and the uneconomical rearing of cockerels from layer lines. We hypothesize that crossbred offspring of local chicken breeds and high-performing broiler or layer hens provide genotypes suited for dual-purpose use. The project RegioHuhn is based on three local chicken breeds, Altsteirer, Bielefelder, and Ramelsloher, taking into account the regionality. Both purebreds and crossbreds were tested for their suitability as dual-purpose chickens. Assessing traits related to welfare and robustness is part of the parallel project ÖkoGen. Crosses provided a variation of genotypes suited for dual-purpose use, with the broiler crosses reaching 55-57% higher meat yield, while layer crosses doubled laying performance compared to local breeds. Animal welfare indicators showed breed-specific variation, which is also reflected in the crosses, e.g. Bielefelder layer crosses showed better feather scores and less keel-bone damage than broiler crosses. Breed-specific differences were found in an infection trial with *Ascaridia galli*, e.g. the Bielefelder had the lowest antibody response and the highest worm burden.

The biodiversity of local poultry breeds: characterisation of two Tuscany breeds to save them from extinction
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The safeguarding and valorization of local poultry breeds may contribute to the sustainability of animal production. The characterization of morphological and productive traits is the preliminary step for the preservation of biodiversity and utilization of marginal lands. The Mugellese chicken is a dwarf breed with a medium neck, broad shoulders, long and horizontal wings, wide, and well-developed breast (especially in the hen) with a typical brooding capacity. On the contrary, the Valdarnese Bianca breed is considered the only original Italian meat-type breed of the national genetic heritage with a long neck, broad shoulders, long and horizontal wings, and long legs and shanks. Both these breeds show frugality, resilience, and resistance to diseases and are particularly suitable for free-range farming. Throughout a 1-year observation, the Mugellese and Valdarnese bianca breeds were characterized for morphological, productive, and reproductive traits and at the end of the trial, for caecal microbial community profile. Data confirmed the morphology of both breeds and their good egg production and growth performances. The main characteristic traits detected were: egg deposition for Mugellese that lay 65.75% of the egg during March–April and the remaining 51.86% in August–September; and growth performance for Valdarnese Bianca that showed a final weight of 1911.33 ± 231.44 g with a feed conversion ratio of 5.62 ± 0.24 . The characterization of the caecal microbial community by high-throughput sequencing of the 16S rRNA gene suggested that the composition of gut microbiota is different in the two breeds.

Session 36

Theatre 4

Multiperformance of slow-growing and dual-purpose strains in organic chicken production: learning from the PPILOW project

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Societal expectations about poultry welfare are increasing, in particular regarding the access of birds to an outdoor run. This possibility to express natural behaviours outdoors can challenge animals in terms of health and welfare, affect animal performance and impact on their environment. Slow-growing strains of chickens are currently used in organic production systems, and dual-purpose strains are now available to avoid the culling of layer male chicks. In the frame of the Poultry and Pig Low-input and Organic production system's Welfare (PPILOW) project, the multiperformance of slow-growing chickens and of males of dual-purpose strains was studied in organic chicken systems. The investigated variables focused on range use, health and welfare indicators, environmental impacts, animal and economic performance, and meat quality. Results showed that a large variability in range use exists, and that many variables can be linked to this indicator. The PPILOW project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement N°816172.

What do dual purpose cockerels feed on in the range area?

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Since the regulation (EU) 2018/848, it is mandatory to provide free range access to cockerels in organic animal husbandry systems. We are investigating different free range systems for their influence on the intake of components of the outdoor run and if age affects the feed preferences by analyzing crop and gizzard contents. In three experimental runs 2022 – 2023, we compared 4 types of range. In the SRC system, cockerels are given access to a strip of poplars with adjacent patches of arable grass (1). In the hedge system, cockerels were given access to an area with a strip of forage hedge and adjacent patches of pastured grassland (2). For each system, control groups were assigned to arable grass (3) or pastured grassland (4) without woody plants. Per run, 400 cockerels (Bresse Gauloise x New Hampshire) were housed in 8 groups of 50 cockerels each. The cockerels were slaughtered at the age of 12, 15 and 18 weeks without fasting and between 2 and 10 birds per slaughter date and group were selected for crop analyses. The crop and gizzard were removed during the regular slaughter process and frozen until further analyses. So far 253 crops and gizzards have been excavated. In animals from pastured grassland, the proportion of longer plant fibers of total stomach content was higher (12 wk: 37.8 ± 11.4 %, 15 wk: 34.7 ± 13.3 %, 18 wk: 29.0 ± 14.2 %) than in cockerels on arable grassland (12 wk: 24.5 ± 10.1 %, 15 wk: 17.1 ± 6.2 %, 18 wk: 11.8 ± 5.4 %). The presence of woody plants did not have an influence therein. Conversely, the proportion of grit was higher in the groups on arable grassland. The variability between individuals was high and requires a high number of samples. The average gizzard content was 33.0 ± 7.8 g (12 wk), 38.6 ± 11.6 g (15 wk) and 42.4 ± 13.3 g (18 wk). The results indicate that dual-purpose cockerels take more than 10 % of their daily feed intake from material of the outdoor run but pastured grassland might require a provision of grit to enhance digestion.

Session 36

Theatre 6

An index for adaptability evaluation of slow-growing chicken genotypes reared in free-range system

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The chickens adaptability to extensive rearing system (ERS) needs the evaluation of natural behaviours, kinetic and foraging attitude, resistance to environmental stress without leaving out economic traits. It should be necessary to find a simple and exhaustive way to define the chicken's adaptability to ERS. With this aim, this research proposed a provisional adaptability index constituted by few and simple variables that may be analysed also in field context. Four hundred Slow-Growing chickens of 4 different strains (Red JA57, Naked Neck, Lohmann Dual and an Italian crossbreed-Robusta Maculata and Sasso hens) were reared in ERS for 81d. In 30 chickens hematological, behavioral, productive and qualitative traits were determined. The 206 variables collected were first reduced to 27, considering the easy of application, cost-effectiveness and scientific value and classified into 4 pillars (behaviors, body condition, meat quality, in vivo health and welfare). Then, an iterative process including discriminant, principal component, and reliability analyses was performed to finally select 6 variables: meat lipid oxidation, carcass yield, feather condition, comfort behaviours, and blood polyunsaturated fatty acids. Results showed that the index can discriminate the different genotypes in terms of adaptability, showing the trends LD>C-B≥NN>RJ, however, the application in the field should be evaluated. This project has received funding from the European Union's Horizon 2020 research and innovation programme under GA n°816172.

Application of Quality of Life approach to evaluate the behavior of four slow growing chicken genotypes reared in free range

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The “Quality of Life” (QOL) represents a new approach that combines the traditional behavioral assessment with the evaluation of the animal’s affective states in a long observation time. Thus, the QOL could be a powerful tool to investigate how animals cope with the environment on the base of the engagement of positive behaviors. The purpose of this study was to compare the behaviors of four slow-growing chicken genotypes reared in free-range conditions by applying the QOL approach. A total of 400 one-day-old chicks belonging to different chicken genotypes (A, NN, CB and LD) were housed in 8 pens with access to pasture area. From 42 to 81 days of age 10 videos/genotype of two hours length were recorded and analyzed by two expert observers using 10-minute sampling intervals. The behavioral variables were classified into positive and negative affective states and indicators of their balance were calculated. Uni- and multivariate statistical techniques were used while the frequency of the behaviors occurrences was categorized by data binning. Results showed that in all the genotypes studied the percentage of positive affective state was higher than the negative ones. However, LD by showing the highest frequency of “sheltering” associated with the lowest frequency of “grass pecking” behavior evidenced a poor interaction with the outdoor area. Nevertheless, the highest frequency of “dust bathing” and “scratching” in the LD genotype balanced the low occurrence of other positive behaviors. Conversely, NN birds were very active and showed the highest occurrences of positive affective states. Further studies are needed to develop a strong QOL methodology to better classified the behaviors into positive and negative affective states and to find a specific indicator to evaluate their balance.

Session 36

Theatre 8

Productive performance and nitrogen excretion of fast-, medium- and slow-growing chicken genotypes fed diets varying in protein-to-energy ratio

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The use of chicken genotypes with slower growth rate for meat purpose has recently gained renewed interest mostly because of animal welfare and product quality reasons. However, limited information is currently available on the impact of nutritional strategies to optimize their productive and sustainability traits. Therefore, a trial was carried out to evaluate the productive performance and nitrogen excretion of fast- (FG), medium- (MG) and slow-growing (SG) chicken genotypes, chosen according to their average daily weight gain (>70, 40-50, and 30-40 g/bird/day, respectively), when fed either a conventional basal diet (CON) formulated to meet the nutritional requirements of the FG genotype, or the same diet with reduced crude protein-to-metabolizable energy ratio (-10% crude protein; LOW), which is representative of commercial diets used for slower-growing chickens. A total of 1,800 one-day-old chicks (600 for each genotype) were allotted in 36 floor pens following a 3 x 2 full-factorial experimental design (i.e. 3 genotypes x 2 dietary treatments; 300 chicks/group). Body weight and feed consumption were recorded on a pen basis at the end of each feeding phase (13 and 27 days) and at slaughtering (35, 56, and 84 days for FG, MG and SG, respectively) to calculate growth performances. At processing, birds were also individually weighed to assess body weight uniformity. Nitrogen excretion was estimated through the indirect method as the difference between nitrogen input and nitrogen body retention. According to the results of a two-way ANOVA, the genotype as well as the administration of LOW diet had a significant effect on performance parameters, particularly on feed conversion ratio. The ongoing evaluation of nitrogen excretion will provide information on the impact of feeding LOW diets to genotypes with different growth potential. Overall, these results can enlarge the knowledge about the effects of nutritional strategies on productive performance and sustainability traits of slower-growing chicken genotypes.

Feather's transcriptome analysis in local chickens fed with two different diets: a possible marker to study lipid metabolism

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Gene expression in feathers is mainly linked to regenerative behavior for physiological needs and in response to trauma. Growth control, morphogenesis and differentiation are the main mechanisms characterizing feathers. Lipid metabolism in chickens is typically studied in liver, abdominal fat, and breast muscle. In this study we analyzed the feathers' expression profile in 60 Bionda Piemontese chickens fed with two different lipid content diets: a low diet (LL, ether extract (EE)=3.6%) and a high diet (HL, EE=9.3%). The aim was to evaluate the possibility in using feathers to study marker genes linked to lipid metabolism. We collected the growing feathers at 150 days of age, 15 males and 15 females/diet. Total RNA was extracted with FastGene® RNA Premium Kit and evaluated for its quantity (Qbit®, RNA Broad-Range Assay Kit) and integrity (RIN, Agilent 2100 Bioanalyzer). 100 ng, RIN ≥ 7 , were sent to AZENTA (Azenta Life Sciences, US) for whole RNA profiling. Different expression analysis was performed using RaNA-Seq software. Results showed a higher expression in the apolipoprotein A1 (APOA1) (p-value = 0.01, log2FC = 1.69) in males fed with HL diet. This was not observed in females. APOA1 is the central constituent of HDL (high-density lipoprotein) and its function is to promote the efflux of cholesterol from peripheral tissues to the liver. The high expression in male chickens fed with HL diet indicates that the excess of cholesterol intake from the diet leads male chickens to overexpress APOA1 to maintain a proper cholesterol homeostasis. In conclusion, this work showed the possibility to use feathers to investigate gene expression in live chicken and identify APOA1 as marker for lipid metabolism study.

Session 36

Poster 10

Suitability of commercial crossbred dual-purpose male chicks for capon production

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The elimination of day-old cockerels of layer breeds poses a problem for the poultry industry. The poultry producers show limited interest in raising male chicks of dual-purpose breeds for meat but these chickens could be used for capon production. The aim of this study was to determine the effect of age and surgical caponization on the selected growth parameters of dual-purpose cockerels. The experiment was performed on 160 crossbred dual-purpose Rosa 1 chickens. The birds were raised to 24 wk of age and fed commercial diets ad libitum. From 16 wk of age, at 4-wk intervals, 8 intact cockerels and 8 capons were selected randomly and slaughtered. At 8 wk and 16, 20 and 24 wk of age, blood samples were collected to determine testosterone levels. The results are presented as means and the SEM. The significance of differences in mean values between age groups was determined by Duncan's test. Average plasma testosterone levels in cockerels reached 0.13 ng/ml at 8 wk of age (castration). Testosterone concentration increased to 2.53 ng/ml in roosters at 24 weeks of age and was significantly higher than in capons (0.15 ng/ml, $P < 0.05$). Final BW weight was affected by caponization and age. Capons had higher BW than cockerels and significant differences were noted in wk 20 ($P < 0.05$) and in wk 24 ($P < 0.05$). Caponization had no effect on the total lean meat content of the carcass ($P = 0.627$), but differences were found between the weights of breast muscles and leg muscles. The weight of breast muscles was higher in capons than in cockerels, 16, 20 and 24 wk ($P < 0.05$). Cockerels had higher leg muscle weight than capons, and significant differences were noted in wk 16 ($P < 0.05$) and in wk 20 ($P < 0.05$). In the present study, caponization had a beneficial influence to an increase in BW or the weights of major parts of muscles. Acknowledgements: Funded by the Minister of Science under „the Regional Initiative of Excellence Program”

Exploring the influence of alternative feed ingredients on meat production and quality in a slow-growing autochthonous chicken breed

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The intensification of poultry farming represents a key issue towards a more sustainable future. Poultry biodiversity is essential in promoting sustainable and high-quality farming systems. This study aims at evaluating meat yields, quality, and color of an autochthonous slow-growing chicken breed, Bianca di Saluzzo (BS), fed with alternative ingredients to replace soybean meal. A total of 96 male BS of 39 days of age were allotted in 12 pens (6 replicates/treatment) and assigned to 2 different feeding programs. The control group (C) was fed a standard commercial corn-soybean meal diet (AME (MJ/kg) 11.8; CP 18.1) while the experimental group (EXP) was fed an experimental diet with the complete replacement of soybean meal with alternative ingredients (pea protein, field bean, barley, maize gluten) (AME (MJ/kg) 11.9; CP 18.1). At 147 and 174 days of age, 12 birds/treatments were slaughtered. Hot and chilled carcass, and organs (spleen, liver, heart), breast and thigh muscles weights were recorded, and the corresponding yields (% live weight) were calculated. Proximate and fatty acids composition was evaluated on the breast meat. Data were analyzed by a Generalized Linear Mixed Model (RStudio, $P \leq 0.05$). No differences were found in slaughtering performance between the two treatments and the two slaughtering age. On the contrary, higher levels of yellowness in both breast and thigh meat were found for the EXP treatment compared to the C group. Finally, the results of proximate and fatty acid composition highlighted no significant differences between the treatments. Overall, the results were comparable between birds fed with alternative ingredients and those on a commercial diet, indicating the feasibility of this approach to promote sustainability of poultry products.

Genetic characterization of Collo Nudo Italiana and Millefiori Piemontese chicken breeds through high-density SNP genotyping

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This study aimed to assess the genetic variability of two indigenous breeds, Collo Nudo Italiana (CNI) and Millefiori Piemontese (MP), and evaluate their variability in the context of Italian local chicken biodiversity. Forty-eight individuals per breed, comprising both males and females, were genotyped through Affymetrix Axiom 600k Chicken Genotyping Array. Both CNI and MP exhibited high expected heterozygosity (0.672 ± 0.117 and 0.565 ± 0.119 , respectively) and observed heterozygosity (0.654 ± 0.139 and 0.601 ± 0.119), and low coefficient of inbreeding (-0.004 ± 0.066 and 0.149 ± 0.405). Multidimensional scaling, phylogenetic tree, and admixture analyses allowed to contextualize CNI and MP with other chicken breeds, including 19 local breeds and 4 commercial hybrids. Results showed that CNI is close to the commercial hybrids, whereas MP is closely associated with Mericanel della Brianza, Bianca di Saluzzo, and Bionda Piemontese local breeds. The present study characterized CNI and MP breeds using high-density chip, also in the framework of Italian chicken biodiversity, and findings are useful for conservation purposes. This work was funded by MASAF (project "TuBAVI-2", PSRN 2014-2022, Sottomisura 10.2).

How does the diversification of farming methods in Europe influence the main indicators of the technological and sensory quality of chicken meat?

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The H2020 INTAQT project (grant agreement n°101000250) is based on the 'One Quality' concept, which encompasses the various facets of intrinsic product quality (nutritional, safety and sensory) as well as the quality of production systems (i.e., economic, environmental and societal sustainability). Its aim is to provide the knowledge and tools needed to characterize, authenticate and improve the quality of products from the diversity of European livestock farms. It focuses on three products – beef, dairy and chicken – which account for around 65% of the economic value of animal products in Europe. Thanks to an extensive network of 90 farms in France, Belgium, Italy, Switzerland and Poland, we have been able to assess the impact of 18 European farming systems on the intrinsic quality of chicken breast and thigh meat. The farming systems studied were both conventional and organic, and covered a continuum of practices from the most intensive to the most extensive, with the use of fast- or slow-growing genotypes (including dual-purpose strains), giving access to more or less enriched environments (including free-range rearing). We propose here to present the first results obtained for the classic indicators of meat quality such as ultimate pH, color, drip and cooking loss, proximal composition, tenderness after cooking and processing yields. These initial results will be supplemented by additional measurements of nutrients or hazardous elements, microbes and shelf life, as well as sensory analysis and consumer testing. Our long-term objective will be to develop multi-criteria quality assessment tools, as well as decision-making tools to guide the evolution towards more sustainable farming practices for the animal, the environment and the consumer.

Session 37

Theatre 1

Getting weaned pigs off to a great start: What we have learned to reduce stress and improve performance

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One of today's biggest challenge in the US swine industry is achieving optimal performance and livability in the post-weaning period. Research from our team has demonstrated that genetics, management, and nutrition all play a role in reducing stress and improving post-weaning performance. Modern genetics that are selected for later maturity in order to improve finishing pig performance exhibit more stress around weaning and have lower post-weaning feed intake and nursery growth performance compared to early-maturing lines. Increased wean age and offering creep feed are pre-weaning strategies that can improve post-weaning performance and livability. Mat feeding and some sensory attractants utilized immediately after weaning have resulted in reduced post-weaning weight loss and nursery removals and mortalities. Sow nutrition plays a role in offspring performance and live yeast probiotics added in lactation diets has resulted in heavier weaning weights and better post-weaning performance. Currently, the US swine industry is able to take advantage of high levels of ZnO in nursery pig diets. However, we continue to conduct research in diets without high ZnO and have identified nutritional strategies that lead to beneficial effects on performance and livability that are similar to those observed when high ZnO is fed. Reducing the acid-binding capacity (ABC-4) of nursery diets by lowering limestone and other ingredients that bind acid as well as adding acidifiers to phase 1 and 2 diets to reach an ABC-4 value of 200 and 250 meq/kg, respectively, has resulted in improved growth performance and livability. Lowering crude protein with the use of feed-grade amino acids can improve fecal dry matter and growth performance can be maintained as long as the SID Lys:CP ratio does not exceed 6.5%. Optimal post-weaning growth performance, livability, and reduced stress can be achieved but a multifaceted approach must be utilized that considers genetic, management, and nutritional influences on health and well-being of pigs.

Effects of early socialisation on growth, health and welfare of piglets raised on commercial farms

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Nowadays, conventional breeding systems are being questioned by public opinion that asks for systems more respectful of animal welfare. Socialisation of suckling piglets might be an appropriate method towards this objective. In this context, we compared the growth, welfare and health of pigs, reared on 5 commercial farms with or without socialisation from the age of 14 days (D14). Frequent visits were made until slaughter to record piglet behaviour and to detect any clinical signs, body and tail lesions. Piglets were weighed on D28 and D40. Salivary cortisol was measured on D28, and hair cortisol on D40 to assess stress levels. Linear mixed models were used to evaluate the effects of socialisation on these variables. According to preliminary data obtained from 4 farms (320 pigs), the socialised piglets showed more exploratory behaviours and positive social interactions than the controls immediately after mixing on D14. After weaning on D28, they still exhibited more frequent positive social behaviours than controls and their body lesions were of lower severity, associated with reduced salivary cortisol concentrations. Nevertheless, we observed lower weight gain during the lactation period for socialised piglets, whose weight on D14 tended to be inferior to the ones of control piglets. On D40 and D68, the frequency of positive social interactions remained higher in the socialised group while control piglets displayed more negative social interactions. Socialisation therefore appears as a possible way of improving piglet welfare although several questions remain especially regarding the impact on the sows.

Spray dried plasma improves piglets' performance and gut function in the first week post-weaning

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Spray dried porcine plasma (SDPP) exhibits potential in reducing the adverse effects of weaning stress in piglets. This study evaluated the efficacy of SDPP to improve performance and gut barrier function in weaning pigs. Ninety animals were randomly distributed into 18 pens (5 pigs/pen; randomised block design with 9 blocks of initial BW; 2 treatments). Between 0-14d post-weaning, piglets were offered pre-starter feed with either soy protein concentrate (62 g/kg; Control) or SDPP (50 g/kg). Between 14-35d all piglets were offered a commercial starter feed. Pigs' individual BW and pen FI were recorded at 0, 7, 14 and 35d of trial. Faecal consistency was assessed using a 5-category score system. At 7d, blood plasma and bile samples were obtained from one piglet per pen for the ELISA quantification of calprotectin, citrulline, and sIgA as biomarkers of inflammation, intestinal function, and mucosal unspecific defence, respectively. Data was analysed by ANOVA using the GLM procedure in SAS. During the first week of trial, SDPP piglets improved FI (215 vs 280 g/d), ADG (132.8 vs 205.3 g/d) and G:F ratio (0.61 vs 0.74) relative to the Control ($P < 0.05$). Over the whole trial, SDPP also improved piglet's ADG and BW. No effect on faecal scores was observed. Calprotectin, sIgA, and citrulline levels were not affected by dietary treatments. It is suggested that the administration of SDPP during the pre-starter phase improves piglet's performance although the specific biomarkers analysed in the current study were not able to explain any beneficial effects on gut barrier function.

Effects of maternal probiotic, and/or piglet dietary tryptophan supplementation pre-weaning, on gene expression in the pre-weaned pigs stomach

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The stomach plays a crucial role both in digestion and in the functioning of the innate immune system and its immaturity poses a significant concern during the post-weaning period. Maternal probiotic supplementation can benefit the sow and also her offspring through maternal transmission. However, little is known about how probiotics may affect the stomach directly or influence its development in young pigs. Tryptophan (Trp), an amino acid known to stimulate acid secretion activity, may potentially enhance stomach function in the pre-weaned pig. A 2×3 study was conducted to investigate the effects of maternal probiotic supplementation, and/or increased piglet Trp on creep intake and on the expression of a panel of genes essential to the functioning of the fundic gland region of the stomach, at weaning. On D85 of gestation, 48 sows were grouped by parity and backfat and assigned to one of two groups: 1) basal or 2) basal + *Bacillus subtilis* and *Bacillus amyloliquefaciens*. On D8 post-farrowing, litters were further divided, and assigned to one of three creep diets resulting in six groups (n=8 litters/group): T1) B17 (basal sow diet+0.17 Trp:lysine creep); T2) B21 (B+0.21 Trp:lysine creep); T3) B25 (B+0.25 Trp:lysine creep); T4) P17 (probiotic sow diet+0.17 Trp:lysine creep); T5) P21 (P+0.21 Trp:lysine creep); T6) P25 (P+0.25 Trp:lysine creep). A total of 48 pigs were sacrificed at weaning (one per litter), and a mucosal sample was collected from the fundic gland region. Gene expression was measured by qPCR and analysed using SAS. Maternal probiotic supplementation tended to increase creep feed intake (P<0.1) and upregulated genes related to acid secretion (ATP4A, CLIC6, HRH2, HDC, CCKBR, CHRM3), enzyme production (CHIA, PGA5), ghrelin production (GHLR) and mucus production (MUC6), in the fundic gland region (P<0.05). Increasing levels of tryptophan in creep feed had no effect on gene expression in the fundic gland region (P>0.05). In conclusion, further research is warranted to determine if maternal probiotic supplementation increased the expression of genes related to acid secretion directly and/or indirectly via increased creep intake.

Session 37

Theatre 5

The leucine-derived gut microbiota metabolite isovalerate enhances the epithelial barrier function in cell monolayers derived from pig ileum organoids

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Introduction: Metabolites produced by the gut microbiota are key regulators of the gut barrier function. The objective of this work was to evaluate the dose-dependent effects of butyrate and branched-chain fatty acids on the epithelial barrier function by using an innovative model of cell monolayers derived from pig ileum organoids. Materials and methods: Monolayers derived from porcine ileum organoids were treated for 48h with butyrate, isovalerate, isobutyrate and 2-methylbutyrate (1, 3 and 5mM). Epithelial barrier function was assessed by transepithelial electrical resistance (TEER) measurement and gene expression was analyzed by qPCR. Results: Butyrate (>1mM) and isovalerate (>3mM) enhanced the epithelial barrier function, as indicated by increased TEER. Butyrate and isovalerate modulated epithelial innate defense system as indicated by the enhanced gene expression of the antimicrobial peptide SLPI and of the cytokine CXCL8 while the expression of the pro-inflammatory enzyme PTGS2 was reduced. Butyrate and isovalerate also upregulated the expression of genes indicating differentiation of enterocytes (FABP6, ARG2) and enteroendocrine cells (CHGA), while reducing the expression of the stem cell makers OLFM4. In contrast, isobutyrate and 2-methylbutyrate had no effect on the epithelial barrier. Conclusion: Our results show that isovalerate, the leucine-derived metabolite, enhances the epithelial barrier function in cell monolayers derived from pig ileum organoids. Therefore, promoting the production of isovalerate by the gut microbiota in vivo could be a novel strategy to promote gut health in pigs.

The impacts of resistant potato starch and *Lactiplantibacillus plantarum* on growth and health of post-weaned pigs

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Lactiplantibacillus plantarum (Lacto) is a probiotic, while resistant potato starch (PS) is a prebiotic that promotes bacterial short chain fatty acid production. This study aimed to examine the effects of supplementing resistant potato starch and *Lactiplantibacillus plantarum*, individually or combined, on the growth and health of weaned pigs. The 96 weaned pigs were sorted into 4 groups and fed for 35 days: (1) control diet (C); (2) C+1% PS; (3) C+2×10⁸ CFU/g of Lacto; (4) C+2×10⁸ CFU/g of Lacto+1% PS (LactoPS). A faecal score (FS) system of 1-5 was used to monitor the incidence of diarrhoea (1=very hard, 5=very loose). There was a Lacto effect (P<0.05) on daily gain (DG) and final body weight (FBW), with Lacto inclusion reducing DG (458 vs 527, SEM 0.017) and FBW (18.6 vs 20.4, SEM 0.437) compared to non-Lacto diets. There was a PS X Lacto interaction on daily feed intake (FI) and FS; the addition of Lacto reduced FI compared to the C diet (764 vs 852, SEM 0.025), but combining Lacto and PS had no effect on FI compared to the PS diet (785 vs 774, SEM 0.025). The addition of PS reduced FS compared to the C diet (2.17 vs 2.50, SEM 0.027), while combining PS and Lacto had no effect on FS compared to the Lacto diet (2.17 vs 2.17, SEM 0.027). There was no effect (P>0.05) of dietary treatment on FCR. Combining *Lactiplantibacillus plantarum* and resistant potato starch had no benefit on growth or faecal scores over individual supplementation. However, potato starch acts as a potential post-weaning anti-diarrhea in post weaned pigs.

Session 37

Theatre 7

Impact of Deoxynivalenol in a Nutritional Calcium Depletion and Repletion Protocol in Piglets

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The contamination with deoxynivalenol (DON) has been shown to modify calcium (Ca) metabolism in pig, but the underlying mechanism remains unclear. This study evaluated the effect of DON contamination, with variable Ca levels in feed, on phosphorus (P) and Ca utilization by pig. A total of 64 piglets (15.7±0.7 kg) received one of 4 treatments during a first 13-day phase: control (DON-) or DON-contaminated treatment (DON+, 2.7 mg/kg) with either a low Ca (Ca-, 0.39%) or normal Ca level (Ca+, 0.65%) with a constant digestible P level (0.40%). During a second 14-day phase, all piglets were fed a similar diet containing 0.65% Ca and 0.35% P without DON contamination. Growth performance, apparent total tract digestibility (ATTD), retention coefficient (RC) of Ca and P and bone mineral content (BMC, with dual-X-ray) of one pig per pen were evaluated. After 13 days of depletion diet in phase 1, DON did not impact growth performance. The BMC was increased in Ca+DON+ piglet, but the opposite was observed, where Ca-DON+ presented the lowest BMC (DON×Ca, P<0.001). The P ATTD was higher for Ca-DON+ piglet (DON×Ca, P<0.01), but their RC of P was lower (DON×Ca, P<0.001). The Ca ATTD was higher for DON+ (P<0.001), but the RC of Ca was higher for Ca-DON- only (DON×Ca, P<0.01). After 14 days of repletion diet in phase 2, the Ca- piglets recovered their BMC deficit in DON-, but not those receiving DON+ (-12%; DON×Ca; P=0.06). The Ca- piglets also had increased Ca (P<0.05) and P (P=0.06) RC. The ATTD of P and Ca in Ca+DON- piglet was lower but was increased in DON+ regardless of Ca level (DON×Ca, P<0.05). Overall, a 13-day Ca deficiency in piglet exposed to DON increased Ca and P digestibility, but not their utilization efficiency, which prevented piglets from recovering their BMC deficit after repletion.

Welfare is the best medicine: Management strategies to prepare piglets for weaning

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Weaning is the most stressful experience in a piglet's life. Traditionally under conventional production systems piglets are simultaneously separated from their mother (and often their siblings), experience a change in diet form and frequency and are moved to a novel environment often with unfamiliar piglets with which they must fight to establish a dominance hierarchy. This occurs at a time when the digestive and immune system are immature which increases the pigs susceptibility to growth checks, disease and antibiotic treatment. In addition to nutritional strategies there are several underutilised management strategies to reduce stress and improve pig welfare. These are characterised by preparing the pigs behaviourally and socially for the transition to the weaner stage and separating the stressors temporally. Firstly, minimising cross fostering, though challenging with large litters, is crucial to ensuring resilient pigs at weaning. EFSA's recent scientific opinion supports the End the Cage Age Initiative to ban the use of farrowing crates for sows and piglets. Free lactation crates promote faster piglet growth to weaning mediated through more space and better opportunities to learn feeding behaviour from the mother. Where sows can 'get-away' from their piglets they can initiate weaning by reducing nursing frequency and thereby accustom piglets to gradual separation from the dam. Co-mingling of suckling litters introduces piglets to unfamiliar conspecifics prior to weaning thereby eliminating their need to fight at weaning and reducing associated injury. It also enables piglets to learn appropriate social skills and to fine tune them at a younger age. Finally, providing piglets with better opportunities to satisfy their behavioural needs to play and explore the environment through provision of environmental enrichment during the suckling period reduces their likelihood of developing damaging behaviours post-weaning. Respecting pigs biological and behavioural evolution ensures good welfare which is the best preventative medicine in addressing performance and health problems for piglets caused by weaning.

Session 37

Theatre 9

The effect of increased energy and amino acid levels in lactation diets on the performance of sows' piglets

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During lactation, it is important for sows to consume sufficient energy for milk production. In the current study, we aimed to investigate the effect of lactation feed type on the growth performance of the sow's piglets. A group of 52 sows was divided over 2 treatment groups: 26 received a standard European feed (9.25 MJ NE, 8 g SID Lys/kg) and 26 a concentrated feed high in energy and essential amino acids (10 MJ NE, 10 g SID Lys/kg) from one week before expected farrowing date until weaning. Sows were fed restrictedly until farrowing. Then feed provision was gradually increased until ad libitum feed intake from d11 in lactation. All piglets were followed until weaning and a selection (10 per sow) until 9 weeks of age. The difference in feed type was reflected in milk composition with lower fat levels (8.1 ± 1.4 vs $8.9 \pm 1.4\%$) and higher urea levels (1012 ± 115 vs 907 ± 118 mg/L) in milk of sows receiving the standard diets. The piglets from the control group tended to be heavier at birth (1.50 ± 0.21 vs 1.35 ± 0.17 kg, $P=0.056$), but no significant effect on weaning weight was observed (7.5 ± 1.2 vs 7.2 ± 0.7 kg $P=0.327$). After weaning, the selection of piglets from the control group showed higher feed intake (537 ± 78 vs 509 ± 87 g/d, $P=0.043$) and faster gain (409 ± 65 vs 386 ± 68 g/d, $P=0.048$) compared to these from the sows receiving the concentrated diets. It can be concluded that no beneficial effect of concentrated lactation diets on the sow's offspring was observed.

Conventional farrowing crates vs. Loose housed system. Effects on performances of sows and piglets

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Housing systems of sows during lactation are change from crates towards a non-restrictive systems, to improve sow welfare, but the loose farrowing systems can increase piglet mortality. In this work we carried out two trials with lactating sows and weaning piglets, 48 + 480 and 44 + 456. The two experimental treatments were the use of Conventional farrowing crates (CV) compared to Loose housing system (LS) where the sow was released 3 days after farrowing. The weight of the sows before farrowing, after farrowing and at weaning (only in the 1st trial) were measured. 24 hours after birth, the litters were placed with 14 piglets. The piglet weight at 24h, weaning (28 days) and at the end of the rearing period (63 days), the dead piglets (crushed or other causes) and the interval between weaning and estrus of sows, fertility and farrowing rates in the next cycle were also registered. The weight of sows did not differ between treatments. Also the piglets weights at diferente ages and the number of weaned piglets did not differ between the two systems. After the opening of the crates, the average number of dead crushing piglets was lower in conventional crates (0.07 vs. 0.35; $P < 0.05$), with no differences in other causes (0.22 vs. 0.39). The average weights of piglets at the beginning and at the end of rearing period, the ADG, the mortality rates and piglets removed did not differ. In the following cycle the interval between weaning and estrus, fertility and farrowing rates did not differ between sows. The farrowing system did not affected the piglets performances but increase their mortality by crushing after the opening of the crates. Funding: This work was also supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Session 37

Poster 11

Effects of uncutting piglets tails on tail biting

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Tail docking is a common practice in almost all the intensive farms of pigs in Portugal and most EC countries. However, with the increasing concern for animal welfare it's no longer possible to perform it routinely as a preventive measure to tail biting. Thus, the main objective of this work was to evaluate the effects of undocked pigs (UP; n=510) and docked pigs (DP; n=510), from weaning to the end of the rearing period and fattening period on tail biting. In the rearing phase the piglets were housed in groups of 17 animals/pen (2.04m x 2.53m; 0.30m²/piglet). In the fattening a total of 953 animals entered the study (480 DP and 473 UP) were housed in groups (19 to 20 animals/pen (3.77m x 4.12m; 0.78 to 0.82m²/animal). The weaning pens had a full slatted floor in plastic and the fattening pens had a full slatted floor in concrete. In both phases, feed and water were distributed ad libitum in a "tube-o-mate" feeder between each 2 pens, with 2 drinkers. Environmental enrichment in weaning pens was achieved by placing a rope with knots and a wooden log. During the fattening phase, in each pen, a drag chain and a two wood logs. During the rearing phase, 21 UP (4.12%) and 0 DP were removed due to biting ($P < 0.0001$). For other reasons, 13 UP (2.55%) and 11 DP (2.16%) were removed ($P = 0.6795$). During the fattening phase, 17 UP due to tail biting (3.59%) and 0 DP were removed ($P < 0.0001$). Due to other causes, 11 UP (2.33%) and 4 DP (0.83%) animals were removed ($P = 0.0642$). In both phases and for all causes, 62 UP (12.16%) and 15 DP animals (2.94%) were removed ($P < 0.0001$). DP did not show any bite outbreak during their lifetime and 68.6% (350) of UP had at least one outbreak, and 34 (9.7%) were bitten a second time. Of the 350 animals affected by tail biting, 236 were victims during the rearing phase (67.4%) and 114 during the fattening phase (32.6%). In the breeding phase, 21/30 pens (70%) and, in the fattening phase, 20/24 pens (83.3%) of UP were affected. This study warns of the problem of abandoning this practice without proper precaution, even controlling the risk factors associated with tail biting as much as possible, the differences between groups throughout life were quite significant.

WelFarmers: The best practices in welfare for pigs

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The WelFarmers project, included in the Horizon Europe Program commenced on the 1st of January 2024 and will be on-going for three years. Pig farmers, research centres and higher education institutions are working together from 8 different EU countries (Spain, France, Ireland, Portugal, Denmark, Finland, Romania and Italy) that represent more than 50% of the pig herd in the EU. This project is setting up eight national innovation networks and four Europe-wide networks of pig farmers, advisors, veterinarians and researchers to address the following challenges; Loose housing of lactating sows Production of pigs with non-docked tails Avoiding pain in castration Space allowance and flooring for growers and finishers The most urgent innovation needs and challenges will be identified in a bottom-up approach and the network will collect and evaluate good practices that meet these needs. The selected best practices will be disseminated through a series of communication and dissemination activities to reach pig farmers in the eight participating countries and in Europe.

Session 38

Theatre 2

Is it possible to end piglet castration forever?

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There is no debate that surgical castration of piglets is unethical and impairs animal welfare. Surgical castration is a painful procedure, with long term negative implications on animal welfare, health, mortality and growth rate. Apart from the higher animal welfare, the feed conversion rate of entire males is higher than castrated by 15-20%. Feeding costs are about 55% of the growing costs of pigs, and therefore are crucial for cost-effective production. In addition, among other reasons, reduced feed intake results in reduced production of greenhouse gas emissions (for example nitrogen excretion is about 15% lower). Still, about 65% of the piglets globally are surgically castrated. The EU has been aiming to ban piglet castration since 2018. The main reason for castration is to prevent boar taint, bad odour of cooked meat, in a small percentage of the pigs. The main obstacle to banning castration and raising entire males is the lack of an efficient on-line automated system for sorting tainted meat at the abattoir. Different methods have been developed and failed to serve as an on- slaughterline solution. To date, there is one at-line method that has been commercialized. The optimal analytical method for boar taint detection needs to meet several requirements. Recently, advanced optics and artificial intelligence showed a promising direction in on-line detection of boar taint. An inexpensive, environmentally friendly solution for the pork industry, might enable ending piglet castration forever

The influence of fibre sources on tail damage in docked pigs

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Dietary fibre is cited as having a role in preventing tail biting in pigs, but greater tail directed behaviour has been reported with increased fibre levels. Rats fed high fibre diets are more anxious and aggressive and have increased circulating lactic acid. Lactic acid should not be the end product of gut fermentation but dysbiosis in fermentation in young animals is more likely and coincides with risk periods for tail biting. We hypothesised that pigs fed diets with increased soluble, fermentable carbohydrates would have higher levels of tail damage. Nine week old, mixed sex pigs (n= 288, 20.3 ± 0.24 kg) were moved into 12 pens and allocated to one of three weaner diets (14 MJ/kg DE); Control- wheat/barley base (3.2% CF, 10.9% NDF, 4.0% ADF), Soy hulls- wheat/soy hulls base (6.0% CF, 14.6% NDF, 4.6% ADF) and Beans- wheat/faba bean base (3.7% CF, 10.8% NDF, 4.9% ADF). On d14, pigs were weighed and inspected for body injuries and tail damage or biting. Data were analysed with sex and diet as fixed effects and pen as a random term. Linear regression was applied to weight data, negative binomial regression to score data, and binary logistic regression to tail damage incidence. There was no significant difference between treatments in weight. Injury score tended to be higher in treatment diets (Soy hulls 1.2 ± 0.10, Beans 1.2 ± 0.10) compared with Control (0.9 ± 0.10; P = 0.087). There was no difference in incidence of tail damage or biting. We did not identify a link between increased dietary fibre and tail biting. However, the trend for higher injury score in the high fibre groups warrants investigation. In future work, faecal and circulating lactic acid will be measured, as well as behavioural tests that quantify anxiety.

Session 38

Theatre 4

Tail lesions monitoring in weaned undocked tail pigs

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This study aimed to evaluate the presence of tail lesions in undocked tail pigs and their evolution from post-weaning to delivery to the fattening farm. More than 60 cycles and 2 thousand weaners were examined across 8 farms, with observations conducted, by the farm veterinarian, at 3 different times post-weaning (7 days, 1 month and 2 months after weaning). The number and the severity of tail lesions have been noted as well as the pigs that were moved to the hospital pens and the losses. The statistical model considered the random effect of the "farm" factor and, in the trend analysis (repeated measures model), the "survey" within-cycle factor. The tail lesions had a peak in the middle of the monitored period, when 17.8% and 7.3% of the 8-week old weaners showed minor and severe injuries, respectively. The animals delivered to the fattening farm were 90.7% of the total original weaners, however, among them, 17.8% were animals recovered from the hospital pens or tail lesioned. The frequency and severity of tail lesions were significantly affected by the farm factor. The results achieved, despite the diversity of the phenomenon in the farms under study, have demonstrated a high incidence of biting in weaners. These findings highlight the importance of the monitoring carried out and the extreme intricacy of the "queue management" problem, especially considering the current production costs and the consequent need to optimise production results while ensuring pig welfare. The study was conducted in collaboration with Associazione Regionale Allevatori of Friuli Venezia Giulia and Veneto.

The effect of stocking density on performance, lesions and soiling

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Providing more space per pig ensures less competition at the feeder, less negative social interactions, and better use of functional space (eat, rest, defecate). This study aimed to evaluate the effect of reduced stocking density on performance, skin and tail lesions and soiling. A total of 177 pigs (Topigs TN70 x Belgian Piétrain) were randomly divided into three treatment groups: a stocking density of 1.3m²/pig (SD1.3, n=14 pens), of 1.0m²/pig (SD1.0, n=12 pens) and 0.8m²/pig (SD0.8, n=10 pens). Both half and fully-slatted pens were included. All pens had a total pen surface of 4.88 m², so the SD1.3, SD1.0, and SD0.8 pens had 4, 5 and 6 fattening pigs, respectively. Sex (barrows and gilts) was balanced within the pen. The trial started at 10 weeks of age (22.6±2.5kg) and ended at slaughter (116.6±12.6kg). Pigs were provided a commercial two-phase diet with a shift at 16 weeks of age. Individual animal weight and feed intake per pen were measured to calculate average daily gain (ADG), feed intake (ADFI) and feed conversion ratio per phase and overall. Carcass quality results were collected at slaughter. Skin and tail lesions as well as soiling of the floor and pigs were scored on 4 time points throughout the fattening period. Performance results and carcass quality were not significantly different between the different stocking densities. The minimal required surface (A) for the pigs at 116.6 kg (W) corresponds with 0.86m² ($A=k*W^{0.667}$, with $k=0.036$ if temperature is below 25°C) (EFSA, 2003), and was thus only compromised for SD0.8 at the end of the fattening period. Pen soiling was significantly higher in the pens with half-slatted floors compared to fully-slatted floors at the last time point, with no significant difference according to stocking density. Skin soiling did not differ over time nor between stocking densities, but was overall higher in half-slatted floor pens.

Session 38

Theatre 6

Effects of light spectrum on health and behaviour of growing-finishing pigs

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Light spectrum was found to affect pig behaviour and ultraviolet B (UVB) light improved vitamin D status, however, it is unclear how these affect welfare. We studied the effects of light spectrum and UVB light on growing-finishing pigs' health and behaviour. Four light spectra were tested; warm white LED light (2594 K), forest daylight LED (4336 K), cool white LED (6235 K) and fluorescent tubes (3787 K). In each spectrum, half of the pigs received UVB light and the other half did not. The health of 224 pigs from 2 batches in 4 rooms of 4 pens was assessed weekly for tail and ear biting lesions, conjunctivitis, tear stains, body lesions, bursitis and lameness. At slaughter, blood was sampled for vitamin D analysis and the presence of post mortem abnormalities on carcasses was checked. Pigs' exploration, social interactions, play and damaging behaviour were observed biweekly for 40 minutes from videos using a 0-1 sampling method. Data were analysed using generalised linear mixed models including light spectrum and UVB and as random effects batch, room, pen, observation week and pig. Effect of light spectrum depended on the week for tear stains, conjunctivitis, body lesions and bursitis (all $p<0.05$). UVB exposure improved vitamin D status ($p<0.001$), and its effect depended on the light spectrum for ear lesions ($p<0.05$). Social behaviour was affected by a UVB and week interaction ($p<0.01$), and the effect of light spectrum on exploration tended to depend on the UVB ($p=0.076$). No light spectrum or UVB effect was found for other health, behaviour and post mortem indicators. These preliminary results show that light spectrum can influence some aspects of pig welfare.

Use of an outdoor run by group-housed sows in the mating area depending on weather conditions

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In 2021, the EU Commission announced its intention to phase out fixation for sows in estrus by 2027, while in Germany the housing standards are currently being changed to group housing. One modification to meet these standards is the addition of an outdoor run in the mating area. Therefore, a total of 263 sows were observed in 15 batches (9 sows/pen) using a camera system (first 4 days after weaning) on a commercial sow farm with a free-access outdoor run. The system continuously recorded the entire area of the outdoor run, allowing it to monitor the sows' visitation patterns. A weather station tracked hourly weather data. First results showed that in a batch with average weather conditions (T: 11.8 °C, hum: 78.4%, rain: 3.1 mm, solar radiation: 153 W/m²), peak visitation times were observed between 07:00 and 17:00, with a high number of visits recorded on the day of weaning ($p < 0.05$). Younger sows showed a higher number of visits compared to older sows ($p < 0.05$). Temperature ($p < 0.001$) and solar radiation ($p < 0.001$) influenced visits, while precipitation showed no significant effect. Further steps are to investigate the impact of extreme weather conditions on sow behavior and outdoor run use to provide insights into optimizing housing conditions for estrus sows. This study is funded by the German Federal Ministry of Food and Agriculture (BMEL) and granted by the Federal Office for Agriculture and Food (BLE; grant number 28N305603).

Session 38

Theatre 8

Study of the attractiveness and longevity of enrichment materials and wood species in a blocked farrowing house and a large gestation group setting

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Two trials were conducted at the IFIP experimental station to provide farmers information to choose suitable equipment. The first trial lasted four weeks was performed in a blocked farrowing house with three groups of 24 sows each. For each group, four solutions were tested: a chewable cylinder, wooden slats fixed to the floor (named octopus), a wooden slat and a natural rubber object fixed in front of the sow. Each week, the objects were rearranged to ensure that all sows in the group were exposed to all four enrichment materials. Behaviour was monitored twice a week using scan sampling for two hours each time, and the rate at which the materials degraded was measured by weighing them. During the observation, the octopus were used 8.9% and the rubber object 10.3%. It is significantly more than the other objects (the chewable cylinder : 3.9% ; wooden in the front of the sow : 5.7%). Additionally, there was greater interindividual variability in the use of the rubber object than that of the octopus. These objects were preferred due to their size, composition, and accessibility, which were better suited for expressing investigative behaviours. The second trial was performed in a dynamic group of 72 pregnant sows to compare the attractiveness and durability of slats made from four wood species: Scots pine, maple, beech, and oak. The trial lasted for three months, during which the duration that sows used each type of slat was measured weekly during a continuous two-hour and a half observation period. The degradation rate of the wood species was assessed based on how frequently the wooden slats needed to be replaced. Scots pine was used on average 80.2% of the observation. It is significantly more than maple (16.1%), beech (20.5%) and oak (19.8%). All objects were mainly chewed. As Scots pine is softer, it degraded more rapidly, which required six times more frequent replacement.

Evaluating the impact of enrichment material in farrowing pens on sow and piglet performance

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Farrowing crates (FC), common in Ireland, are used to protect against piglet mortality and ease handling, but limit the sows' expression of natural behaviour due to restricted movement and often minimal enrichment. Loose manipulable material could provide an outlet for nesting behaviour prior to farrowing, and investigatory behaviour during lactation, improving sow welfare. However, it is important that any associated gut fill does not lower sow feed intake, which could have negative effects on milk yield, back-fat, and piglet performance. This study investigated the impact of providing three loose materials to sows in conventional FC; Straw (n = 12), Haylage (n = 14), and Hay (n = 14), and a control (Rope; n = 14). Sow performance, feed intake, and tear stain scores, and piglet average daily gain (ADG) and weaning weights were evaluated. Loose material was provided via a metal box attached to the side of the feed trough for oral extraction, and refilled twice daily if at least half empty. In the Rope pens, a hessian rope was hung on the side of the FC, within easy reach of the sow's mouth. Fifty-four healthy sows, balanced by weight, backfat and parity, were assigned to a treatment one day prior to entering the farrowing room. Sow feed intake was recorded daily during lactation, and locomotion and back-fat scores taken at weaning. Tear stain scores were taken prior to farrowing, one week post-farrowing, and at weaning. Piglets were weighed at birth and weaning to calculate ADG. None of the measurements taken were affected by treatment. Thus providing loose material in the farrowing pens throughout lactation did not appear to inhibit feed intake due to gut fill, or impair sow or piglet performance.

Session 38

Theatre 10

Positive effect of nesting material on crushing losses in free farrowing pens with short-term fixation

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Aim of this study was to investigate the effect of organic nesting material on piglet crushing. The study was carried out on a farm in Northern Germany where farrowing systems with a short-term fixation (2 days a.p. until approx. day 7 p.p.) and a pen area of 7.0 m² were installed. In 11 batches, the sows were provided with straw (n = 67) as nesting material in a permanently accessible rack from gestation day 112 until birth. Jute bags were offered in the control pens (n = 71). Results show that the straw group has significantly lower total crushing losses and crushing losses within the first three days p.p. compared to the control group (jute bags: 7.37 %/6.15 %, straw: 5.91 %/3.34 %). It is also striking that in the straw group only about half of the crushing losses occurred within the first three days p.p., whereas in the control group almost all crushing losses happened during this period. These data suggest that offering straw influences the event of crushing losses, resulting in a continuously decreasing risk of crushing. This is probably the result of a change in the behavior of the sows in connection with the supply of straw, which is currently being verified using video recordings. This work is funded by the German Federal Ministry of Food and Agriculture (BMEL) based on a decision of the Parliament of the Federal Republic of Germany, granted by the Federal Office for Agriculture and Food (BLE; grant number 28N305603)

Can black soldier fly live larvae improve postweaned piglets' welfare?

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We tested the effects of black soldier fly (BSF) live larvae administration on some growth and welfare indicators of weaned piglets. Forty-eight piglets were distributed in pens of 6 and allotted either to control (C, kept in standard commercial conditions) or to live larvae group (LL, receiving 300-600g/pen/day of BSF LL as a supplement). The trial lasted 40 days. Growth parameters were recorded and behaviour was video recorded every 10 days. Body and tail lesions were assessed using the Welfare Quality protocol and hair cortisol was quantified as a chronic stress indicator. Data were submitted to ANOVA using the group as a fixed effect and the pig as a random effect. Average daily gain was 0.53 in C and 0.52 kg in LL, average lesion and tail scores were 0.58 and 0.12 in C, and 0.23 and 0.00 in LL ($P > 0.05$). Video analysis showed that LL piglets spent 14% of the observed time interacting with larvae, reducing time spent at the feeder (16.6 vs. 21.9%, $P < 0.001$), interacting (neutral interactions 5.1 vs. 8.7%, $P < 0.01$; aggressive interactions 2.3 vs 3.5%, $P < 0.05$), manipulating the enrichment (hanging wooden or chains, 1.2 vs. 2.3%, $P < 0.05$) and biting the tail of their pen mates (0.0 vs. 0.5%, $P < 0.05$) compared to C. Hair cortisol was tendentially lower in LL (8.1 vs. 10.9 pg/mg, $P = 0.051$). Pending more extensive behavioural observations, our results indicate that BSF LL administration can improve animal welfare by shifting the exploratory behaviour from the body of pen mates and pen equipment to larvae. No significant effect in terms of lesion score reduction was observed, but hair cortisol showed tendentially lower chronic stress levels. Supported by EU grant n°J83C22000830005-AGRITTECH research programme.

Session 38

Poster 12

Effects of deck height during transport of weaners: in-vehicle conditions and clinical indicators

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We aimed to examine if deck height affected in-vehicle conditions (temperature and CO₂ concentration as indicators of ventilation) when weaners are transported at stocking densities corresponding to EU-regulation, and to examine consequences in terms of mortality, weight loss, and post-transport skin lesions for pigs at 20-25 kg. We performed two field trials consisting of 32 semi-controlled journeys under conditions modelling commercial Danish practice; half of 8 and 23 h, respectively, run in warmer and colder periods from June 2021 to March 2022. Deck heights of 60 vs 80 cm (trailer) and 70 vs 90 cm (truck) were compared. We collected a mixture of sensor time series, GPS records, weather data from a public online source, clinical examination of focal animals, and various information on settings like opening of ventilation shutters and animal stocking density. Data were analysed by (generalised) linear mixed effects models. Lower deck height had only sporadic and minor effects on the indicators examined. Horizontal differences in temperature and CO₂ concentration were detected among vehicle compartments. In the colder period, large differences were found between study sensors placed close to the pigs and embedded temperature sensors placed in the outer wall of the vehicle. Thus, the livestock driver may be misinformed by information from embedded sensors and left with limited chances of adjusting conditions; being examined further by a follow-up study.

Determination of social interactions in a group of gestating sows based on location, posture and orientation
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To study social interactions, it is necessary to identify individuals, their posture and their relative proximity. More precisely, the orientation of the two protagonists in relation to each other through 3-point vectorization (snout, neck, tail) provides information about the valence of the interaction (positive or negative). A study was carried out to assess the value of this approach for detecting positive and negative social interactions. Two pens of 13 gestating sows were studied during two consecutive gestations, with 3 cameras installed above each pen. For each pen and gestation, videos collected from 00:00 (feeding time) to 02:00, on days 30 and 103 were manually analysed. The number of interactions and their valence (positive vs. negative) were collected continuously, as well as the location in the pen, the posture of both sows involved and their orientation during the interaction (snout-snout, snout-tail...). The valence of the interaction was analysed using a generalized linear mixed-effect model with location, orientation and posture of both sows as fixed effects. Positive interactions were mainly observed in resting areas ($P < 0.001$) and negative interactions mainly in the feeding area ($P < 0.05$). Positive interactions were more of snout-snout and snout-tail (sniffing the ano-genital area) types, and negative interactions were more of head-head and head-body types ($P < 0.001$). Recipient sows were more lying during positive interactions than during negative ($P < 0.01$). These results suggest that variables such as location, orientation and posture provide useful information about the valence of interactions. Work is currently underway to develop automatic detection of these parameters using image analysis.

Strategies for minimizing dust in straw-based-systems for pigs
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Dust concentrations in straw-based systems for pigs repeatedly cause respiratory problems due to increased dust levels. To determine the reduction potential, an automated litter and dust removal technology was tested in a multi-year trial in a functionally separated animal welfare stable for fattening pigs. The values of reducing dust allow conclusions about the good effectiveness of straw dedusting using a Strohmatic Welfare system as well as the additional dosage of an oil-containing substance using a low-pressure spray system. Through extensive monitoring over a period of 1.5 years, seasonal trends were recorded. The daily course of the dust mass concentration and the number of dust particles is in any case influenced by the activity of the animals as well as the frequency and type of littering processes. Particle counts tended to be higher in fall/winter (reduced air rate, air moisture content), with statistical significance in terms of season and particle counts. The number of particles was clearly distinguishable under different measuring conditions (manual litter with/without oil, automated litter and dedusting with/without oil, automated litter without dedusting). To test a new type of straw dedusting (Strohmatic Light) and the influence of oil spraying on dust pollution (AirCleaner low-pressure oil spray system) in the room air, a comparison of variants over several days was carried out in a follow-up measurement in a dislocated stable building for fattening pigs. The excellent effect of the oil application was once again confirmed. In summary, the avoidance and binding of dust is seen as an indispensable component in the lying area. In terms of labor management, innovation and automation in the animal sector, as well as to mitigate quality defects in bedding materials, the best available technology is clearly a mixture of oily substances and simultaneous automation of the bedding process to prevent respiratory diseases caused by particulate matter in humans and animals.

Building sustainable systems at the era of farm-to-fork and agroecology: Unraveling the interplay of quality attributes of animal-source foods

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The quality of animal-source foods includes safety, commercial, sensory, nutritional, technological, convenience attributes and societal image. The latter, also referred to as extrinsic quality, covers ethical, cultural and environmental dimensions related to animal farming and product's processing. All these attributes can be improved, but also impaired, along the value chain from the farm (breed/genotype, farming system and practices) through slaughtering and processing conditions, up to consumption. Some antagonisms between quality attributes have been identified in pork, lamb and poultry. For example, grazing is favorable to image and lamb meat nutritional quality, but increases the risk of off-flavors. Solutions are sought to overcome these tensions. In organic pig farming, feeding can help solve the issue between animal welfare (avoiding males' castration) and sensory quality (reducing boar taint risk). There are also synergies between farming practices. For instance, combining breeding and feeding strategy improves sensory and nutritional quality of pork, while reducing its environmental impact through the relocation of feed resources. The interactions between animal genotype, farming and processing practices are also the root for the high sensory quality and typicality of Geographical Indications products. Quality of animal-source foods is thus part of, and a lever towards sustainable livestock systems. Quantitative assessments of antagonisms and/or synergies among quality attributes, using multi-criteria approaches from a Global Quality perspective, are pivotal to identify pathways for sustainable systems.

Session 39

Theatre 2

Dairy Milk vs Plant-Based Milk Alternatives: Exploring Fat Nutritional Indices in Retail Products

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While milk remains a dietary staple, the popularity of plant-based milk alternatives (PBMA) is increasing. Limited data on nutrient profile at retail level and few comparative studies prompted research on nutritional indices in dairy milk (cow and goat) and PBMA. Samples included 7 cow milk, 8 goat milk, and 14 PBMA (7 almond, 4 oat with seeds/nuts, 1 rice with coconut, 1 pistachio). Fatty acid composition was assessed as follows; polyunsaturated to saturated fatty acids ratio (PUFA/SFA), atherogenicity index (AI), thrombogenicity index (TI), and hypocholesterolemic to hypercholesterolemic fatty acid ratio (h/H). Differences between groups were determined using ANOVA test. Significant differences ($P > 0.001$) among the groups (cow, goat, PBMA) were observed across all indices. PUFA/SFA ratio exceeded the recommended value of 0.45 in PBMA (1.86) whereas it was very low in cow and goat milk (0.06 for both). PBMA exhibited lower AI (0.56) compared to dairy milk, which approached the recommended value of 3 (2.66 for cow, 2.45 for goat). Similarly, PBMA had lower TI (0.48) compared to dairy milk, which surpassed the recommended value of 3 (3.55 for cow, 3.23 for goat). The h/h ratio was notably higher in PBMA (8.45), while cow and goat milk showed lower values (0.55 and 0.62, respectively). Despite better nutritional indices, ongoing research is crucial as the commercial variety of PBMA is continuously expanding. Additionally, a comprehensive assessment of the nutritional value of PBMA necessitates research on parameters such as mineral and vitamin content, as well as anti-nutrient factors.

Case study of the market situation of calves from Alpine dairy farms and the effect of dams' grazing during the last three months of gestation on auction parameters

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The main aim of the study was to assess the recent evolution of cattle breeding in Alpine areas based on the number, quality and price of calves sold at local auctions in the South Tyrol region over the last decade, as case study. In addition, the effect of grazing during dam's late gestation on calves' auction quality was investigated to assess whether the inclusion of pasture in Alpine production system has an impact on auction parameters of calves beside its well-known positive effects on cattle welfare and the environment. Similarly, to other geographical areas, the study revealed that the numbers of crossbred calves (especially from specialised dairy breeds) has increased over the last years also in the Alpine area, because of more targeted breeding plans with an intensive use of beef bulls' semen. Further, purebred calves from dual-purpose breeds (Simmental, Alpine Grey, Pinzgauer) received a higher remuneration in terms of price and market value at auction than purebred calves from specialised Holstein Friesian and Brown Swiss cows. A part of these animals could be available to support an innovative alpine meat supply chain which, if grass-fed, could further strengthen the current landscape maintenance service provided by the mountain livestock husbandry. Dams grazing during the last 90 days of gestation had a limited negative effect on calves' liveweight and quality traits at auctions.

Session 39

Theatre 4

Truly superior? Consumer ratings of meat from an endangered German pig breed

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The endangered German Angler Saddleback pig (AS) is said to have a high palatability of the meat for its increased carcass fatness as compared to modern genotypes while scientific evidence is low. Therefore, this study sought to reveal consumers' appreciation of AS meat versus more commercially (CO) used breed types. All pigs were fattened at a performance testing station until an average slaughter weight of 102 kg. AS had outdoor access, received a restricted diet reduced in energy and protein, and were offered silage daily. CO had no outdoor access and received a standard diet. Upon electrical stunning, pigs were slaughtered and loin muscles of eight pigs per breed type were dissected 24 hours post mortem. A total of 81 consumers each received two products of both AS and CO to rate their liking of appearance, taste and overall liking using nine-point hedonic scales. Samples were served monadically in a blocked, randomized order. First, 4×4 cm pieces of only lean meat (LM) of both breed types were served followed by steaks (SK) with adjacent subcutaneous fat standardized to about 1 cm. Samples were fried to a core temperature of 70 °C using contact grills. On average, overall liking was higher for CO than for AS ($p=0.01$). Appearance of LM was rated better than SK ($p<0.01$). In contrast, taste liking was neither affected by breed type, type of presentation nor their interaction. Overall, these results do not support that AS meat is unanimously superior. Consumers' depreciation of visible fat is considered a marketing challenge for fatty breeds like AS. Additional analyses that also account for consumer demographics and attitudes towards pork might further explain these preliminary findings.

Informing about higher levels of animal welfare increases sensory consumer liking of pork

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This double-blind consumer (n=300) study confirms that, labeling livestock husbandry levels positively affects consumers' sensory perception even when the meat is physically identical and experiment-related confounding factors are ruled out. Herein, loin steaks were obtained from pigs (same breed, same diet) raised under different conditions according to the German husbandry level system (haltungsform.de) HL1 (conventional) vs. HL3 (outdoor access). Four within-subject conditions were randomly tested: a blind-tasting of HL1 vs. HL3 (control pure, CP), a blind-tasting with both products being HL3 (control identical, CI), an informed-tasting with correctly labeled HL1- and HL3-products (control original, CO), and an informed-tasting with both products being HL3 but labeled as HL1 vs. HL3 (placebo, PL). Prior to tasting, participants received information on pig husbandry levels to ensure a common level of knowledge. Liking was recorded for various sensory attributes (appearance, odor, taste, palatability, aftertaste, overall impression) using 9-point hedonic scales. In result, both blind tastings (CP and CI) revealed no significant difference in any attribute indicating similarity between meat from different husbandry levels when undisclosed. Yet, when the actual husbandry level of the meat being tasted was informed about (CO), consumer liking for the HL3-meat compared to the HL1-meat was higher ($p \leq .001$) with the difference potentially caused by factual product differences. While the placebo condition (PL) eliminated this confound the higher ($p \leq .001$) liking of HL3-(vs. HL1) meat persisted. This marketing placebo effect of labeling livestock farming conditions on consumer liking of meat is potentially moderated by individual morality. Overall, the findings demonstrate the importance of credence quality attributes such as livestock farming conditions and animal welfare for consumers' perception of quality. Hence, husbandry systems ensuring higher levels of animal welfare are considered a chance for increased consumer appreciation of animal products.

Session 39

Theatre 6

Implementing sustainability frameworks on product level- exploring the usability

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Social and environmental sustainability are inherently complex and interconnected. They involve multiple dimensions, such as economic development, social equity, human rights, environmental conservation, and cultural preservation. Developing a framework that adequately addresses the diverse and interrelated aspects of both social and environmental sustainability can be challenging. Several methodologies for assessing both environmental and social sustainability exist such as SAFA, INSPIA, SAI for farm level. The product level frameworks have not been implemented in practice on a large scale and have not been extensively evaluated. Hence our aim was to explore the usability of two relevant frameworks adaptable on product level. The framework selection was based on multiple selection criteria, including the integration of environmental and social metrics and proven real-world application. Subsequently, these frameworks were applied in case studies of Norwegian milk and Norwegian greenhouse-grown tomatoes. Our findings highlight the introduction of new biodiversity-related metrics and the incorporation of new social considerations. However, challenges related to context-specificity and cultural diversity as data availability and quality emerged as crucial factors in assessing food sustainability at the product level. Additionally, our findings underscore the necessity for a common scale or standard to facilitate comparisons among food products within and across categories.

Economic sustainability and resilience of extensiveness in fattening pig farms

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The modern consumer expects more and more quality food animal origin, which they generally associate with extensive farming systems. This perception exists for all species, but it is particularly strong for pigs, which in most cases are housed in intensive farming systems. Within the mEATquality project, innovative strategies were developed to meet new societal demands, environmental requirements and economic needs of entrepreneurs. The project proposes and follows a step-by-step analysis strategy considering the extensiveness factors of pig production as a key point. These data were collected from conventional, free-range and organic farms and processed using standardised sustainability assessment protocols. Data collected from farms in Denmark, Italy, Poland and Spain provided a general description of the farm, highlighted key environmental and animal welfare issues and provided economic and productivity information on the farm. The economic protocol was useful to collect fundamental information to understand the economic performance and its variability according to the different degree of extensiveness of pig farms. To this end, a set of economic indicators was calculated, such as production costs, feed costs per head, farm income per labour unit, returns on capital and technical efficiency of the production process. In addition, the economic resilience of the pig farms was assessed, based on specific qualitative indicators, designed to describe the 'power' of the entrepreneur within the value chain, his entrepreneurial skills, training and innovation willingness and the resilience of farm resources.

Session 39

Theatre 8

Green willow biomass as feed source for pigs

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Background: Farmers integrate trees such as willows (*Salix* spp.) into outdoor pig systems to enhance animal welfare, carbon sequestration, nutrient capture, and biodiversity. However, the potential for these trees to support feed self-sufficiency has been overlooked. While previous research has demonstrated the nutritional advantages of green willow biomass for ruminants, its suitability for pigs has not been thoroughly investigated. Objective: To investigate green willow biomass as a feed source for fattening pigs and gestating sows. Methodology: Willows were coppiced during the dormant season, and the regrowth of shoots with leaves were either a) directly foraged by fattening pigs as fresh matter from May to July or b) harvested in June, ensiled, and subsequently fed to gestating sows during the following winter season. Green willow biomasses were analyzed for dry matter (DM), metabolic energy (ME), crude protein (CP), and amino acid content (including lysine and methionine). Additionally, pigs' performance (e.g. live weight and back fat gain) and voluntary intake of willow biomass were assessed. Results: The fresh and ensiled green willow biomass showed DM contents ranging from 21% to 28%. CP content (of DM) varied from 13% in silage (a mixture of leaves and shoots harvested in June) to 26% in leaves harvested in May. Preliminary findings suggest high palatability of willow biomass when foraged directly or harvested/ensiled early in the growing season (May-June). Conclusion: Green willow biomass is a potential protein source for pigs. Future research should prioritize strategies aimed at exploiting the multifunctional role of trees within outdoor pig systems while supporting farm feed self-sufficiency.

Organic pig farming: from feeding strategy to meat organoleptic quality

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Pig diet is one of the major factors from farm to fork determining pork quality attributes, including organoleptic traits (appearance, texture, flavour and odour). An experiment was conducted with 77 organic uncastrated male pigs (Piétrain x Large White) in two batches in time, each divided in two groups. One group received a control diet (C) corresponding to the organic specifications. The other group received a test diet (Bio+), mainly based on French raw materials, containing more fibre (Faba bean) and omega-3 fatty acids (linseed, camelina) and had access to forage. Each group was reared in one pen in the same building on deep straw bedding with free outdoor access and fed ad libitum until slaughter. Meat was analysed by a trained sensory panel and by SPME-GC-MS to identify the main odor-active volatile compounds. Meat was lighter ($P < 0.05$) and had a more intense red colour ($P < 0.1$) in Bio+ than in C pigs. Meat from Bio+ pigs had a lower aromatic persistence ($P < 0.05$), whereas odour intensity, tenderness and juiciness did not significantly differ between the groups. None of the samples were qualified as boar tainted by the panellists. Identified volatile compounds belonged to aldehydes, hydrocarbons, alcohols and ketones. Of the 29 compounds identified, 10 had a prevalence of over 50%, with the same proportion in C and Bio+ pigs. Only two compounds, benzaldehyde (almond odour) and heptanal (fatty odour) were found in all samples. Finally, the Bio+ diet influenced some sensory properties but did not modify the odour, flavour or aromatic profile of pork. A joint analysis of the volatile profile and sensory data is planned as part of the global-quality approach.

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Theatre 10

Land use efficiency to protein production of French dairy cattle contrasted in their feeding systems

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Livestock production occupies 56% of agricultural land in France and raises the question of efficient land use, as a component of environmental sustainability. The Land Use Ratio (LUR) is the ratio of the potential for plant protein production on land used for livestock production, both on-farm and off-farm for purchased feed, to the animal protein produced on the same land. A LUR of less than 1 indicates that animal production is more efficient than crop production. Two ways of expressing the quality of plant and animal protein in the LUR calculation have been proposed: 1) using total digestible protein (van Zanten et al., 2016), 2) using the edible fraction of plant and animal proteins and their respective digestible indispensable amino acid score (DIAAS) (Hennessy et al., 2021). The LUR was applied to 12 case studies of French dairy farms with different feeding systems: grass-based, mixed and maize-based. Animal protein production was compared with a 6-year crop rotation: wheat, maize, faba bean, rapeseed, wheat and lupin. Grass-based systems use more land per kg of protein produced than mixed and maize-based systems (78.6 ± 13.6 , 53.0 ± 11.4 and 33.6 ± 5.4 m².kg⁻¹, respectively). However, because they use a high proportion of non-arable grassland (66% of the total area used), grass-based dairy systems are more efficient for protein production than mixed and maize-based systems (LUR according to van Zanten of 0.91 ± 0.62 , 2.69 ± 1.37 and 2.29 ± 0.63 , respectively; LUR according to Hennessy of 0.37 ± 0.26 , 0.98 ± 0.46 and 0.81 ± 0.22 , respectively). Dairy systems with a high proportion of non-arable grassland are more efficient at producing human-usable protein than crops (LUR < 1 according to both methods of calculations). Moreover, considering the quality of proteins through the edible fraction and the DIAAS score, which indicates the suitability of the amino acid profile of proteins, significantly reduces LUR values of dairy cattle systems (LUR < 1 according Hennessy) and therefore improves their land use efficiency assessment. This may influence the comparison between animals and crops in sustainability assessment.

Characterization of small egg producers in the O'Higgins region, Chile

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A survey was conducted to evaluate the perception of egg quality and small egg production farms in the O'Higgins region. This survey involved 91 farmers and included 27 questions in three sections: i. Sociodemographic characteristics, ii. Farms characteristics, and iii. Egg quality. The analyses involved an ANOVA test, the comparison of means for quantitative variables, frequency tables, and Chi2 tests for qualitative variables. Among the farmers, 81.3% were women with an average age of 57±12 years. No significant differences were found between the ages of women and men. No significant differences in the educational level were found due to gender; however, a relationship was observed with age ($p < 0.001$), indicating that younger farmers had a higher percentage of professional education. On average, farms maintained 186±280 layers, with 54.4% having fewer than 100 laying hens. Daily production averaged 136±230 eggs, with a laying percentage of 77.9±46.6%. Regarding marketing, 40% of producers exclusively sold at home, while 72% made additional sales at free fairs, warehouses, local markets, or supermarkets. Hens were housed in cages in 4.3% of the farms, while 78.5% used balanced rations, 31% employed temperature control, and 49.5% used light management. Size was the most used variable to classify eggs, followed by freshness. Farmers prioritized characteristics associated with egg quality, including shell cleanliness, egg flavor, and freshness, while yolk color was less important. The prevalence of female producers and their relatively advanced age should be taken into account when considering policies and activities for the agricultural development of egg production in the region.

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Farm level economic effects of introducing forage crop silage in the feed regimen to fattening pigs

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Introducing biomass from locally produced rotational grass-clover by adopting an alternative feed strategy in pig diets reduce the climate impact and have the potential to improve farm profitability. However, the size of this potential in practical pig farming is not yet well studied. The economic effects of including forage crops in the pig diets of a typical conventional and organic pig production in Sweden was therefore evaluated. A farm simulation model was built for assessing the farm level economic effect of including forage crop silage in the pig diets as part of a total mixed ration (TMR) in a typical conventional and organic pig farm in Sweden. A partial budgeting framework with stochastic elements was used to analyse the farmers' decision-making on the farm income. Results showed that despite an observed increase in investment and operational cost, farmers attain some increment in net income in both the conventional and organic systems. The increase in net income was 13% and 34% higher than the increase in investment and operational cost for including silage in the conventional and organic systems respectively and was associated with extra income obtained for high lean meat content, reduced cereal feed intake and surplus silage sold. Giving these potential economic gains, investments in the conventional and organic pig farming system could be expected since there is private economic reason for farmers. Reflecting the high initial investment costs for reconstructing or adapting the building and feeding system to fit the new feeding regime, investment support from public policy for the reconstruction of pig housing needs to be considered.

Evaluation of milking attitude and milk quality in native Ciuta sheep breed

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The Ciuta sheep is a small, local breed native to the Central Italian Alps. While primarily raised for meat production, there is historical documentation of its traditional use for cheese production. This aspect could provide valuable insights into the sustainable utilization of this local breed in Alpine farms. Consequently, this study aimed to characterize for the first time the quality of Ciuta's milk. The experiment was conducted at one sheep farm located in Valtellina, Lombardy. A flock comprising 11 ewes and their lambs was utilized and housed in a single barn equipped with a catch feeder. All ewes were subjected to the same management system, involving lambing in late December-January followed by lamb suckling until the initiation of the trial (February), when ewes were at 49 ± 8 days of lactation on average. The ewes were manually milked once a day in the morning, and following the morning milking, lambs were allowed to remain with their mothers until late afternoon. Throughout the day, the flock grazed on native pasture, while during the night, they were housed in shaded open pens. Additionally, the ewes were provided with local hay ad libitum and supplemented with 200g/sheep/day concentrate during milking. Sheep behaviour was recorded during milking sessions twice a week, aligning with the days designated for milk sample collection. Furthermore, udder conformation scores, encompassing udder suspension, udder height, and teat placement, were documented for each ewe. Milk production was recorded and individual samples of milk was dispatched to the accredited laboratories of ARAL (Regional Association of Farmers) for compositional analysis, conducted in accordance with official methods (ISO 9622:2013, IDF141). Additionally, the fatty acid profile of milk samples was determined by gas chromatography in our laboratory. Preliminary findings revealed average values of 3.9g/100g for fat, 5.5g/100g for protein (including 4.1g/100g of casein), 4.8g/100g for lactose, 91.7×10^3 somatic cell count (SCS), and 35.6 mg/dl for urea.

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Poster 14

Milk production in Rubia Gallega suckler cows: estimation methods and correlation with morphological and anatomical measurements

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Milk production in Rubia Gallega suckler cows: estimation methods and correlation with morphological and anatomical measurements Moreno¹, T., Montes¹, C., Crecente¹, S. ¹Centro Investigaciones Agrarias de Mabe-gondo-AGACAL. Apto 10.15080 A Coruña *teresa.moreno.lopez@xunta.es The selection of cows with good maternal aptitude, such as adequate milk production, should be one of the priority lines in the development of beef herds with suckler cows in pasture-based production systems, in order to obtain calves with proper fat deposition and unique meat quality characteristics. The milk production was measured in Rubia Gallega cows using two methods (weigh-suckle-weigh: WSW and mechanical milking: MM) during their lactation period (from 115-185 days of calves' age) and while they were on pasture supplemented with grass and corn silage from 1st-March-15th April, and after that on exclusive pasture. In WSW and MM methods, calves were separated from the dams the day before, remaining without suckling for 18 hours. At the WSW method, calves were weighed before, then reunited with their mothers for 30 min for suckling, and weighed again afterward. For MM method, a portable milking machine was used. The milk production is expressed in kilograms from the liters measured in each milking, using a conversion factor of 1.030 (BOE n°332) for weight to volume. The highest milk production was recorded in April and early May (6.43kg MM and 5.58 kg WSW, p-value=0,07), coinciding with calving intervals between 140 and 170 days, and the lowest were recorded in late May and June (5.95kg MM and 5.38kg w-s-w, p-value=0,15). Both methods showed a similar production estimation. The better correlations between the milk production and the morphological measurement, zoometric index and udder measurements were with the bistiliac breadth (0,54MM and 0,44WSW), dactyl-thoracic index (0,55MM and 0,72WSW), and teat caliber (0,43MM and 0,3WSW), respectively (confidence interval 95%, Pearson's correlation test).

Total phenolic content and antioxidant profile – A comparative study between retail goat and cow milk
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Milk contains essential nutrients like proteins, calcium, fatty acids, vitamins, and bioactive compounds such as antioxidants. However, detailed information on the antioxidant capacity of milk is still lacking. The study investigated differences in the total phenolic content (TPC) and antioxidant profile between cow and goat retail fluid milk produced in Greece. Full fat samples from conventionally produced cow (n=36) and goat (n=36) milk were collected monthly for one year from three major food retailers. Both cow and goat samples were sourced from the same manufacturers. Milk samples had a seven-day shelf life, and the pasteurization procedure was standardized to prevent variations in the antioxidant profile resulting from interactions between antioxidants and different industrial heat-treatment procedures. TPC, Free radical scavenging activity (DPPH), Reducing Power Activity (FRAP) and Reducing Power Activity (ABTS) were determined. Differences between milk types were assessed with independent T-tests. Highly significant differences ($P < 0.001-0.01$) were observed between the two types of milk in all examined parameters, with higher values found in cow milk compared to goat milk. Most profound differences were found in TPC (0.64 vs 0.36 mg GAE/mL in cow and goat milk respectively). The findings are related to differences in the feeding regime and husbandry practices between cow and goat dairy production systems. Support: “AGROTOUR” Project (MIS 5047196), “Reinforcement of the Research and Innovation Infrastructure”, NSRF 2014-2020.

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Poster 16

Valorisation of the triple attitude in Ciuta sheep through the characterization of productions: the VAL3CIUTA project

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The VAL3CIUTA project aims to enhance the triple purpose (milk, meat, wool) of the Ciuta sheep, an autochthonous breed originating from the Valtellina and Alto Lario regions, through the characterization of its productions. This breed represents the Italian smallest sheep breed in the Alpine area, being robust, agile, and frugal, particularly suited for life in a rustic habitat. It is predominantly used for meat production, but historical evidence suggests traditional use of Ciuta sheep milk and wool in the past for cheese production and garment manufacturing. Ciuta sheep are considered at a severe risk of extinction. In recent years, projects for the protection of the breed have been initiated, resulting in approximately 400 officially registered individuals in the breed registry. The breed recognition has provided a driving force for the territory of origin to promote its products, ensuring a comprehensive added value to supply chain project, also linked to the ecosystem services associated with this type of product. The starting hypothesis of the VAL3CIUTA project is that enhancing the breeding triple purpose, coupled with the promotion of local genetic resources and traditional derivative products, can add value to local animal husbandry, thus supporting the conservation of this local breed. This, in turn, contributes to the conservation of the landscape and biodiversity, aligning with the multifunctionality principle of agriculture. The project involves two Departments of University of Milan and three farms located in the Valtellina region, which have been involved for years in the protection of the Alpine territory and endangered fauna, particularly focusing on the recovery of local autochthonous breeds.

Dietary supplementation of orange peel ingredient in lactating ewes: effect on yoghurt physicochemical characteristics

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Animal feed production is increasingly favoring efficient use of alternative feed sources, with significant potential for incorporating food industry by-products. The study investigated the impact of supplementing ewes' diet with dried ingredients produced from orange peels, a primary by-product of orange juice production, on the physicochemical traits of yoghurt. Ewes were divided into groups receiving either processed (enzymatically hydrolyzed) or unprocessed orange peels (11% DM inclusion level) (groups POP and UOP, respectively), while the control group (CON) followed a standard diet. Three batches of yogurt were prepared and on storage day 14, characteristics like colour (chroma & whiteness), syneresis, viscosity and hardness were evaluated. ANOVA test was used to compare groups. Supplementation of ewes' diets with dried orange peel affected chroma ($P < 0.05$) and firmness ($P < 0.001$). Yoghurts from the UOP treatment, displayed lower colour saturation and firmer texture compared to POP and CON groups, with no effect observed on the other characteristics. Enzymatically hydrolyzed dried orange peel ingredients can be effectively used in the diet of lactating ewes. Funding: EU PRIMA Program for Research, Technological Development & Demonstration-grant agreement n°2013/ (NEWFEED, <https://newfeed-prima.eu/>).

Dietary supplementation of orange peel ingredient in lactating ewes: effect on yoghurt sensory characteristics

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Nowadays, animal feed production is increasingly favoring efficient use of alternative feedstuffs with significant potential for utilizing food industry by-products. The present study assessed the effect of dietary supplementation with dried ingredients produced from orange peels, a by-product of orange juice production, on the sensory characteristics of sheep yoghurt. Milk was collected from ewes receiving either processed (enzymatically hydrolyzed) or unprocessed orange peels (inclusion level 11% DM) (groups POP and UOP respectively) while the control group (CON) was fed a conventional diet. Three batches of yoghurt were evaluated by taste panelists, who frequently consumed sheep yoghurt. Parameters like overall appearance, colour, aroma intensity, aroma, taste intensity, taste, acidity, texture (mouth & spoon), aftertaste, and overall acceptability were assessed using a 7-point hedonic scale. ANOVA test was used to compare groups. In all groups, the scores for all parameters surpassed the acceptability threshold (score = 4), with no differences ($P > 0.05$) among treatments. In conclusion, supplementation of ewes' diet with dried orange peel, either processed or unprocessed, had no negative impact on yogurt's sensory characteristics. Funding: EU PRIMA Program for Research, Technological Development & Demonstration-grant agreement n°2013/ (NEWFEED, <https://newfeed-prima.eu/>).

Discrimination of organic goat retail milk by fatty acid biomarkers

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Consumer interest in organic goat milk is growing, yet authenticating organic products poses challenges. The study examined differences between retail organic and conventional milk by the application of fatty acid biomarkers for authenticating organic milk. Conventionally and organically produced milk samples from the same dairy company (n=3) were collected monthly from May 2019 to April 2020. The content of individual fatty acids/lipid classes, including pentadecanoic (C15:0), margaric (C17:0), vaccenic (C18:1 trans 11), conjugated linoleic acid (CLA) (C18:2 c9, t11), linoleic (C18:2 n-6), α -linolenic (C18:3 n-3), and n-3 fatty acids, was examined. The effect of production system, season and the interaction between production system and season within each company was evaluated with nested design ANOVA. There was no effect ($P>0.05$) of production system in all biomarkers whereas the effect of season ($P<0.05$) was observed in C15:0, C18:1 trans 11, C18:3 n-3 and n-3 fatty acids. Regarding the interaction between production system and season significant effects ($P<0.05$) were mainly observed for one of the three companies and particularly in winter. Fatty acids affected were C18:3 n-3, CLA and n-3 fatty acids. Although, there were no differences in production system, variations in particular acids between brands in certain seasons might be related to differences in the goat management practices employed in particular regions. Acknowledgement: "AGROTOUR" Project (MIS 5047196), NSRF 2014-2020.

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Poster 20

Application of NIR spectroscopy coupled with DA-PCA for the authentication of lamb meat

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Rearing systems, linked to animal welfare and nutraceutical characteristics, are novel meat features that can drive consumer choice. The need to develop a traceability and authentication meat system emerges, especially for niche sectors such as local lamb meat. The present study aims to test the application of discriminant analysis based on principal components to near-infrared spectra derived from the meat of lambs reared with different feeding systems. Meat samples derived from Massese lamb, slaughtered at 69 days, reared in three conditions: extensive system linked to only pasture diet (n= 70), indoor system associated with concentrate and hay feed regime (n=74) and semi-extensive system linked to pasture, concentrate and hay diet (n=50). Samples of lamb meat referred to three muscles (Longissimus dorsi Triceps brachii and Semimembranosus) were acquired by Antaris II FT-NIRS (Thermo Fisher Scientific). Samples were scanned as intact and fresh meat considering the infrared region (3999 to 9999 cm⁻¹), and 32 scans were performed for each one. After, a multivariate exploration of original spectra was applied on 3112 wavenumbers by discriminant analysis of principal components (DAPC) using the factextra R package. A cross-validation was applied for class assignment where the full dataset was used for model training, and the discriminant functions were extracted based on all samples. The first two components explain 98% of the total variability of the dataset (75.3 PC1 and 22.6 PC2 as %of variance). The DAPC model resulted in an overall assignment success rate of 81,9% for rearing systems, using two discriminant functions.

Characterization and typology of egg consumers in the Central zone of Chile

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An online survey was conducted to characterize egg consumers in the Central area of Chile with the aim of defining consumer groups. The study utilized frequency tables and a multivariate analysis comprising four stages: 1. variable selection, 2. Multiple Correspondence Analysis (MCA), 3. Cluster Analysis (CA), and 4. Discriminant Analysis (DA). A total of 415 surveys were collected, generating 134 variables from 38 questions. Variations in purchasing and consumption behavior were observed across demographics such as age, gender, occupation, rural residence, family size, and income level. After variable selection, 48 variables were included in the MCA, revealing three dimensions that explained 71% of the data variance with a Cronbach's Alpha coefficient of 0.9. The CA identified four distinct consumer groups representing 8.3%, 60.4%, 22.8%, and 8.5% of the respondents, respectively. The DA achieved a classification accuracy of 93.9%. Notable differences among groups included gender and income level, while factors influencing purchasing and consumption behavior encompassed reasons for egg consumption, frequency of consumption, and importance placed on various attributes such as refrigeration, price, shell color, cleaning, packaging, shell condition, size, brand, and certification. Additionally, concerns regarding nutritional intake, familiarity with concepts like free-range farming and carbon footprint, and quality indicators such as yolk and white appearance and egg size were identified. The findings underscore the multifaceted nature of consumer behavior, influenced by socio-economic factors as well as levels of awareness regarding sustainable production practices and animal welfare.

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Poster 22

The effect of gender on meat quality of the longissimus lumborum muscle in growing-finishing pigs

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Pig producers consistently strive to improve production efficiency and profitability, as well as to adapt production systems to the binding rules and regulations, and increasing consumer expectations. Meat quality depends on a number of factors, which can be divided into genetic and environmental. Gender can significantly alter the meat quality of fattening pigs. The aim of this study was to evaluate the effect of sex on meat quality in growing-finishing pigs. Meat samples were collected from gilts, entre male pigs, surgically castrated males and immunocastrated males. Surgical castration was carried out at 5 days of age, whereas immunocastration was performed with two injections of Improvac administered 4 weeks apart, at 9 and 15 weeks of age. Gender was used as the source of selection. Nourished with complete feed mixtures. 4-phase feeding was used. Feed and water were available ad libitum. Pigs of each group were kept in separate pens, and they were slaughtered at BW of around 107kg surgically castrated males to 117kg entre male pigs. The pigs were slaughtered according to the usual procedure in meat plants. Samples of the longissimus lumborum muscle (musculus longissimus lumborum, LL), approximately 10 cm thick, were collected from the right half-carasses to determine the proximate chemical composition, physicochemical properties and sensory attributes of meat. The content of dry matter, total protein (Kjeldahl method), crude fat (Soxhlet extraction) and crude ash. Gender had no influence on the quality or processing suitability of pork, but it affected intramuscular fat content, which is an important consideration for consumers. Funded by the Minister of Science under "the Regional Initiative of Excellence Program".

Effect of sex on the quality and fatty acid profile of the longis muscle in danbred hybrid pigs

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The aim of the study was to check the effect of sex on individual quality and fatty acid (FA) profile in DanBred hybrid pigs. Longissimus dorsi (LD) muscle samples were collected from gilts and entire male pigs during the fattening period, they were kept separately in the pens of group animals. Gender was used as the source of selection. Nourished with complete feed mixtures. 4-phase feeding was used. Feed and water were available ad libitum. The slaughter was carried out in accordance with applicable laws. Longissimus dorsi muscle samples were taken with the half-carcass from the lumbar section between the 3rd and 4th vertebrae. The pH₂₄ meter was measured with a pH-meter and the meat color was measured in the CIE L*a*b system. Contains: dry matter, total protein – Kjeldahl method, raw material – Soxhlet method and ash of origin (AOAC, 2007) and FA profile of origin. The fatty acids were separated by gas chromatography. Fatty acid methyl esters (FAME) according to various Peisker methods. Fatty acids identified by improving their retention times with pure FAME standards (Sigma-Aldrich, St. Louis, Missouri, USA) and peaks in the analyzed samples. The degree of acidification does not differ by gender. Higher content of meat of origin for gilts. Longissimus dorsi muscle samples from gilts were characterized by a higher content of chemical components. The source of origin and high statistical significance between the sexes of the animals and the fatty acid profile were found. Funded by the Minister of Science under “the Regional Initiative of Excellence Program”.

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Poster 24

Comparison of the slaughter weight, slaughter characteristics and carcass composition of South African Boer Goats and Dohne Merino sheep

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Sheep production holds significant importance in South Africa's small stock industry, whereas goat products are predominantly distributed in the informal sector. This study aimed to assess and compare the impact of species and gender on carcass composition, meat quality, and fat depth in Boer goats and Dohne Merino sheep of identical age and upbringing. Upon reaching 120 days of age, the kids and lambs were weaned and introduced to a feedlot environment up to 12 months of age when they were slaughtered. There was no significant difference observed between the slaughter and cold carcass weights of Boer goats (63.8kg; 32.4kg) and Dohne Merinos (61.6kg; 30.9 kg). However, the slaughter and cold carcass weights of the castrated males (65.3kg; 33.2kg) were notably higher than those of the females (60.1kg; 30.1kg) ($P=0.01$). The dressing percentages for Dohne Merinos (50.1%) and Boer goats (49.5%) fell within the expected range of 42.7% to 55.4% for sheep and goats. Boer goat castrates exhibited a dressing percentage (51.0%) similar to that of Dohne Merino wethers (50.0%), Boer goat females (47.9%), and Dohne Merino ewes (49.9%). Dohne Merinos (8.3mm) deposited significantly more fat on the longissimus lumborum compared to Boer goats (4.6mm) ($P<0.01$) at the same age, attributed to the earlier maturation of Dohne Merinos and distinct fat deposition patterns. Results from the study, based on three-rib cuts, indicated notable differences in lean muscle yield and fat and bone content between sheep and goats. Goat meat may be an alternative food option for consumers who prefer meat with a lower fat content.

Beef × dairy crossbreeding in mountain farms: major effects on carcass weight and retail cuts

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For this study, the data collected from 3443 crossbred heifers slaughtered weekly from September 2019 to December 2023 were provided by the Breeders Federation of the Autonomous Province of Trento (North of Italy). The heifers were sired by Belgian Blue (BB) bulls on dams of different dairy breeds: Brown Swiss (BS), Holstein Friesian (HF), Simmental (Si), Alpine Grey (AG), Rendena (Re), and Crossbreds (Cr). Heifers were born in 441 dairy farms, weaned in 7 specialized “weaner” farms and fattened in the same intensive fattening farm. Data for each heifer concerned weaning and fattening length, slaughter age, the weight of carcass (CW) and of 17 main retail cuts, and the average daily carcass gain (ADGc). Retail cuts weights were expressed quantitatively in absolute weight and qualitatively as incidence on CW. All the data were analyzed with a mixed model including the dam breed and slaughter season as fixed effects, while birth and weaning farm and slaughter date within season as random. The results highlighted, with small differences in weaning and fattening lengths, the better performances of the BB×Si compared to the other breed combinations for carcass and retail cuts weights and ADGc. Expressing the data qualitatively, the Alpine breeds (Re and AG) had higher incidence of the principal cuts over the entire carcass, indicating lower losses and/or trimming cuts used for hamburger production. These findings renew the interest, also in mountain areas, for the beef × dairy meat production to improve the economic sustainability of dairy farms. The study is part of the INTAQT project funded by the European Union’s Horizon 2020 research and innovation programme (grant agreement N°101000250).

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Poster 26

Rheological and protein thermal denaturation properties of eggs from different farming systems

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This study aimed to evaluate rheological characteristics and protein denaturation of eggs obtained from three different types of laying poultry farms, intensive, dual purpose and extensive. The flow curves of both egg whites and whole eggs, assessed by rotational rheometry, revealed a shear thinning behavior. Raw egg whites were more viscous than whole eggs due to the higher protein content of the former. Lower steady shear viscosity was recorded for the egg whites obtained from the intensive system, while the variability of values for the extensive system was higher. Differential scanning calorimetry showed no differences among the farming systems in lysozyme and ovalbumin denaturation temperatures of the egg white detected at ~ 65 and 80°C, respectively. The temperature and enthalpy values of an endothermic transition at ~ 82°C of the egg yolk samples, was ascribed to the composite mixture of lipids, lipoproteins and contaminant egg white proteins; these responses were similar among the farming systems, whereas the enthalpy values had higher variability for eggs of the extensive system. Overall, the farming system appeared to affect the variability rather than mean values of the rheological and thermal parameters, with samples from the extensive system exhibiting higher heterogeneity in their physical properties. However, due to the limited number of farms involved in the study and several substantial confounding factors involved (e.g. breed, feed ration), the results should be interpreted cautiously. Funded by EU (HORIZON H2020) under the acronym CodeReFarm Grant No: 101000216

Some genomic insights into the spatio-temporal dynamics of domestication and dispersal of sheep

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The transition from the Pleistocene to the Holocene is characterised by strong global warming, which forced plants, animals and humans to adapt quickly to the new environmental conditions. Human adaptation led to a new way of life and the domestication of plants and animals. A better understanding of neolithisation could be helpful in addressing the current phase of dramatic ecological restructuring and species extinction. To investigate the spatio-temporal dynamics of domestication and the evolution of small ruminant husbandry during and after the agricultural revolution, we combine zooarchaeological studies with palaeogenomics and population genomics of ancient and contemporary domestic and wild sheep and goats. During the screening phase of the ancient samples, we obtained 76 complete mtDNA genomes from the Neolithic sites Aşıklı Höyük (75) and Göbekli Tepe (1). In addition, there are 26 complete mtDNA genomes from the Chalcolithic site of Güvercinkaya. To test alternative demographic hypotheses, we combine the above genomes with hundreds of genomes from contemporary domestic and wild sheep across Eurasia, as well as ancient genomes contributed by other groups. We clearly show that domestication itself does not lead to a genetic bottleneck, but that this only occurs after domestication in the course of western expansion into Europe and Africa. We also clearly show that the entire genetic diversity of today's domestic sheep can be explained by a single domestication centre. The zoo-archaeological findings in combination with genomics clearly confirm the reproduction of small ruminants directly within Neolithic settlements such as Aşıklı Höyük and refute the recently proposed "catch-and-grow" strategy as well as some older theories.

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Theatre 2

What does the past tell us about the future – examinations of remains from domestic livestock

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Identifying animal species in zooarchaeology, i.e. distinguishing between sheep and goat or domestic and wild animals, can be a challenge. Thus, an inventory of precisely addressed animal skeletal remains is essential as a basis for determining unknown animal bone material. Furthermore, this unique collection enables scientists to shape and evaluate their research tools in zooarchaeology. The comprehensive collection of domestic animal skeletons at the university Halle-Wittenberg, once generated from farm animals and their wild forms over a period of more than 100 years, ideally fits this purpose. The skeletal material with its accompanying data covering various time periods and breeds reflects a wide variation of domestic livestock in mainly six major species (pig, horse, cattle, sheep, goat, dog). Manfred Teichert, former curator of this collection, was one of the founders of zooarchaeological research in Germany. Jointly with Boessneck and Müller, a method for distinguishing sheep and goat bones was published (1964). Today, as geomorphometry (GMM) has replaced two-dimensional descriptions as a spatial method, the Halle collection of animal remains is used to evaluate and expand zooarchaeological as well as livestock research. As an example, third lower molars from modern specimens readily distinguish sheep from goats using GMM. Nowadays, the transferability of this method to archaeological samples has been demonstrated (Jeanjean et al., 2023). In addition, 3D-scans of skulls and postcranial bones made available online in a repository will open a new window for zooarchaeological research worldwide.

From Wild to Tamed and Back: demographic reconstruction of Sardinian sheep from whole-genome sequences

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Currently, three sympatric sheep (*Ovis aries*) lineages are found on Sardinia: the Sardinian mouflon, thought to represent the feral remnants of the earliest domesticated sheep, the Pecora Nera di Arbus (hereafter Nera), a local breed showing ancestral features, and the Sarda sheep, strongly selected for milk production. Despite their cultural and agricultural significance, the evolutionary history of these three sheep lineages is still unclear. To unravel the evolutionary origins of the Sardinian sheep, we generated whole-genome sequences of 19 Sardinian mouflons, 6 Sarda and 9 Nera sheep, and co-analyzed them with a dataset of over 30 wild and domesticated sheep lineages. We explored the population structure, built a phylogenetic tree, and investigated the ancestry components. Results confirmed production-driven introgression in Sarda and suggested a first translocation of the Sardinian mouflon lineage followed by a later introduction of the domestic lineage originating Sarda and Nera sheep. Performing demographic reconstruction, we confirmed the time of emergence of the early domestic lineage at ~12,000 YA, from which the most modern domesticated sheep split around ~6,700 YA. Finally, we dated the split of Sarda sourcing from the Nera lineage around ~4,800 YA. Further analysis will reveal potential gene flow between the three lineages and allow the detection of genes involved in local adaptation and agronomic traits.

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Theatre 4

Differential trajectories of the annual reproductive cycle in sheep and cattle from Neolithic to modern times
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The regulation of fertility is an important aspect of reproductive control in farm animals, which have inherited vestiges of seasonal reproduction from their wild ancestors. Cattle and sheep seem to have followed different paths in this respect. Like the current representatives of their wild ancestor the oriental mouflon, modern sheep under temperate latitudes express a seasonal anoestrus. This has considerable consequences for flock management and product availability. However, out-of-season lambing is possible, particularly in breeds with less profound anoestrus – typically Mediterranean breeds. In contrast, while the last representatives of their wild ancestor, the aurochs, were described as seasonal breeders in the 16th century Poland; and while feral cattle populations in Europe reproduce seasonally, today's domestic cattle are non-seasonal breeders. The history of these two trajectories is still poorly understood, but recent methodological advances in zooarchaeology allow to go back to their origins. The seasonal cycles recorded in tooth enamel during growth can be reconstructed from oxygen isotope ratios, making it possible to determine the birth seasons of animals of the past. We will present an overview of the recent results which, on the one hand, demonstrate the existence of autumn lambing as early as the 6th millennium BCE in the western Mediterranean and, on the other hand, the persistence of highly seasonal reproduction in cattle until at least the 4th millennium in continental Europe. The reasons for these different trajectories will be tentatively explained and discussed.

African indigenous cattle genealogy revealed by whole-genome tree sequences

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Africa has a uniquely rich cattle diversity with about 150 known cattle breeds or populations comprised of *Bos indicus*, *Bos taurus*, and their crosses. Various studies investigated their diversity and adaptation. However, methods that efficiently capture genetic differences and evolution between such populations are lacking. Here, we built whole-genome ancestral recombination graphs encoded via tree sequences to describe the genomic evolution of 193 African indigenous cattle from 15 breeds – Baoule, Gourounsi, Kuri, Logone, Boran, Ghanaian shorthorn, Guinean N'dama and Nigerian N'dama from *Bos taurus* origin; Zebu Gobra, Zebu Maure, Baggara, Butana and Kenana from *Bos indicus* origin; and one *Bos indicus* X *Bos taurus*, Djakore. Our results revealed the unique diversity, genomic evolution and subspecies differentiation of African indigenous cattle. The findings from this study will pave the way for future biological discovery and more informed breeding especially in admixed cattle populations.

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Theatre 6

Unravelling the genetic diversity among Nubian, Old English, and Anglo-Nubian goat breeds

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In order to evaluate the genetic diversity and structure among Nubian goats and understand their relationships with Old English, Anglo-Nubian and worldwide goat breeds, 6,006 animals were studied. Among them, 558 goats were genotyped by using 60K chip, and additional 5,448 genotype goats using 52K chips goats were obtained from collaborators and publicly available data. In this study, genomic regions under positive selection were explored. Nucleotide diversity, heterozygosity, and excess of homozygosity were used to estimate the genetic diversity and genomic inbreeding. Breed relationships were assessed through the Fixation index (FST) and principal component analysis (PCA). The average nucleotide diversity for Nubian goats ranged from 0.00075% in the population from Egypt to 0.00083% in the population from Algeria. Nubian goats from Algeria showed the highest observed heterozygosity (38.0%), while Anglo-Nubian goats displayed the lowest (26.3%). The pattern was reversed for the excess of homozygosity. Pairwise FST-values between goat populations ranged from 0.02 (Nubian from Sudan vs. Nubian from Ethiopia) to 0.28 (Anglo-Nubian vs. Old English). The PCA revealed distinct clusters of goat breeds from Africa, Europe and Asia. Nubian goats clustered with African goats. Interestingly, Anglo-Nubian goats were located outside the African cluster, bridging clusters between Africa, Asia, and Europe. Results from runs of homozygosity (RoH) islands analysis support the presence of discernible traits such as adaptation, innate immune response, heat tolerance, reproduction, and pigmentation in Nubian goats. Further research will focus on identified regions and subsequent gene discovery.

Genetic diversity of eight local Belgian sheep breeds using pedigree and SNP data

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Conserving genetic diversity in local livestock breeds is vital to guarantee their future survival. Genetic diversity is a prerequisite for adaptability to environmental changes and resistance to emerging diseases, becoming more pronounced with climate change. We investigated the genetic diversity of eight indigenous sheep breeds in Belgium, based on pedigree and single nucleotide polymorphism (SNP) analysis. In total, more than 600 sheep were genotyped using the GGP Ovine 50K array. Effective population size (N_e) was estimated to be low for all screened sheep breeds, in both pedigree- ($N_e = 17$ to 90) and SNP-based ($N_e < 24$) computation methods. With a minimum sustainable population threshold of 100 animals according to FAO, all eight screened Belgian sheep breeds are considered endangered. Pedigree-based inbreeding estimates were between 3.1% and 11.3% whereas ROH-based inbreeding estimates ranged from 4.1% to 8.6%. In the next stage, 28 different ROH islands were identified for the studied breeds and compared with ROH islands found in international sheep breeds. Interestingly, common ROH islands were found between populations that are close either geographically or in terms of breeding goals. Some of these regions were mapped to known genes associated with specific traits, but further analysis is required to investigate the potential origins of ancestral haplotypes or the underlying genes in these (possible) selection signatures. In summary, this study provides the first genetic characterization of the local Belgian sheep populations, confirming their endangered status and contributing valuable information to the global dataset of genotyped sheep breeds.

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Theatre 8

Integrating zooarchaeological, historical and genomic data to explore potentials for conserving the indigenous cattle breed of Cyprus

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ARETI is a multidisciplinary research project that weaves together evidence from different fields and disciplines, including zooarchaeology, ancient and contemporary cattle genomics, iconography, history and folklore studies, to explore human-cattle interactions on the island of Cyprus. Focusing primarily on cattle bone assemblages retrieved from several archaeological sites across the island, spanning the early Neolithic to Roman times, ARETI investigates the impact of historically significant events, including human migrations, agricultural intensification, population expansion, urbanisation and climate change, on cattle management practices and genetic structure. To explore further cattle genetic admixtures, population structure and the timing of *Bos indicus* introduction on the island, we analysed archaeological cattle bone samples at the Smurfit Institute of Genetics (TCD), while nasal swab samples from 90 unrelated cattle belonging to the island's modern indigenous breed have been collected and analysed at the Cyprus Agricultural Research Institute. Beyond presenting the project's main scientific results, our presentation will also demonstrate how public outreach activities, including the design of several educational activities for children, the production of a documentary and the collaboration with local craftspeople for the creation of products inspired by the thousands of years of history and tradition surrounding the local breed, can further highlight the local cow breed's cultural value and contribute to its conservation.

Evolution of genetic parameters for production traits and conception rate in the Dutch Holstein population
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Limited population size and selection can reduce the genetic variances of traits compared to base population genetic variances. These changes are a result of drift and the Bulmer effect, and can also impact the genetic correlations between traits. Here, we aim to evaluate the evolution of genetic parameters for production traits and conception rate (CR) in the Dutch Holstein population since 1990. The data included around 855,000 305 days-in-milk records for fat, protein and milk yields (MY), and around 676,000 records for CR at first artificial insemination for first-parity Holstein cows. Using a Gibbs sampling approach, changes in genetic (co)variances due to drift and selection were estimated by sex and per year of birth with bivariate models (one production trait and CR), in addition to base population (co)variances at five generations before 1990. For cows born in 1990, genetic variances were 22% lower for MY and 12% lower for CR than the base population genetic variances. From 1990, genetic variances for cows increased on average by 0.3% (of the base population genetic variance) per year for MY, and decreased by 0.2% per year for CR. Corresponding genetic covariances were rather stable over time. Finally, genetic correlations between MY and CR changed from -0.45 in 1990 to -0.41 in 2016. Similar trends were observed for sires, but with steeper increases for genetic variances of production traits and correlations, especially from 2010. This project has received funding from the European Union's Horizon 2020 Programme for Research & Innovation under grant agreement n°101000226.

Session 40

Theatre 10

Genetic diversity and population structure of the endangered Tanzanian Mpwapwa cattle breed

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Mpwapwa cattle is a synthetic dual-purpose breed developed at the Mpwapwa Cattle Research Station, Tanzania, in the 1920s. The development and maintenance of Mpwapwa cattle have faced many challenges, and this breed has never been characterized at the genomic level. Our objectives were to assess the current genetic diversity and population structure of Mpwapwa cattle owing to its highly admixed origin and decline in active genetic management in recent years. Hair samples were collected from 251 cattle from different agro-ecological zones in Tanzania and were genotyped with the Bovine 100K SNP chip (Neogen Geneseek®). After quality control (using PLINK 1.9), we assessed heterozygosity (PLINK 1.9), inbreeding (detectRUNS package in R), and admixture (LEA package in R). We found the observed heterozygosity (0.32) was higher than the expected based on observed allele frequencies (0.29). Furthermore, more than 75% of studied animals had a runs-of-homozygosity-based inbreeding higher than 20%. Admixture analyses have indicated separation of the sampled animals by agro-ecological zones. Our results provide information for developing conservation and improvement strategies for this endangered Tanzania cattle breed.

Genomic analysis of Greek mouflon

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Six (6) blood samples were obtained from an equal number of mouflons that were hunted during the 2022 hunting season (November – December 2022) on the Sapienza Island (Methoni, Peloponnesse) of Greece. Genotyping of the samples using the 50K SNP array followed. Application of standard quality criteria resulted in a final number of 32,263 SNPs used for further genetic analyses. Levels of heterozygosity and homozygosity (via autozygous DNA segments i.e. ROHs) were estimated, while principal components, admixture and phylogenetic analysis were also performed using additional genotypes from other mouflon populations (Asiatic, n=13, Cyprus, n=3, Sardinian, n=23, and Corsica, n=11) and a typical domesticated breed of sheep (Chios, n=23) as an outgroup. In the Greek mouflon samples, the mean observed heterozygosity was low (mean=0.215, SD=0.006), while three of the six individuals were found to have a degree of consanguinity between 2nd (half siblings) and 3rd degree (first cousins). The inbreeding rate of Greek mouflon individuals ranged from 20.5% to 29.8% (average= 25.0%, SD=3.5%) with 70% to 100% of the observed inbreeding arising from relatives matings occurring in the distant and middle past (time horizon beyond 10 past generations). Based on the results of the analysis of principal components, admixture and phylogeny, the specific samples of the Greek mouflon were genetically classified between those of Corsica and Sardinian mouflons.

Session 40

Poster 12

Morphostructural and phaneroptic variability of creole goats in the Southern Highlands of Peru

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This study aimed to characterize the morphostructural and phaneroptic variability of creole goats in the Southern Highlands of Peru. A total of 269 females older than two years, without signs of crossbreeding with specialized breeds, were used from Ica (three districts) and Ayacucho (four districts), Peru, located from 2500 to 3800 masl. Six phaneroptic (color, beard, horns, wattles, number of teats, teat direction) and four morphometric (weight, abdominal perimeter, body length, thoracic perimeter) characteristics were recorded. Factorial analysis of mixed data (FAMD) was applied, followed by a hierarchical classification analysis, using the package FactoMineR, R software (version 4.3.2). Six orthogonal principal components were found, and the first two explain 41% of the total variability observed in the study. The analysis revealed three types of creole goats that were mainly differentiated by the presence/absence of horns and beards, and the number of teats. Type 1 included mainly goats from Ayacucho, with composite coat colors, horns, and supernumerary teats. Type 2 was determined by goats with beards, without wattles, and with the correct number of teats. Type 3 was characterized by being from Ica, without beards or wattles, and with parallel teats. It is concluded that in the Southern Highlands of Peru, there were three types of creole goats with some characteristics in common, such as their large thoracic capacity and a tendency to develop dual-purpose (milk and meat) production. The variability observed represents an opportunity to establish policies that allow conservation and genetic improvement through a selection of this valuable zoogenetic resource.

Genetic analysis of Mediterranean wild rabbit populations (*Oryctolagus cuniculus*) using whole genome sequencing data

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Abstract Genetic analysis using whole genome sequencing data of 8 wild rabbits sampled from 4 Mediterranean islands (Lemnos, Schinoussa, Thira and Malta) and 4 individuals of domesticated rabbit breeds (Rex and New Zealand) was carried out. The analysis involved numerous steps (applying base sequence quality criteria, mapping sequences to the species reference genome (latest version: GCA_009806435.2_NW_1.0), identifying single nucleotide polymorphisms (SNPs), etc.) and was performed using various Bioinformatics tools available on the Galaxy web platform (usegalaxy.org). Estimation of the percentage of heterozygous loci as well as principal components, admixture and phylogeny analysis, followed. The percentage of heterozygous loci ranged from 43.6% (Schinoussa1) to 58.7% (NZ1) with the domesticated breeds (Rex and NZ) showing higher heterozygosity rates when contrasted to the Mediterranean wild rabbit populations. Principal and admixture analysis showed genetic differentiation of the domesticated rabbit from the wild rabbits of Mediterranean origin. This differentiation was also confirmed by the phylogenetic analysis in which the samples from Lemnos were classified in a common subclade with sampled from the Malta island.

Session 41

Theatre 1

Estimated output from assisted reproductive techniques (OPU/IVF) in Holstein cattle associated with the genetic background

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By improving all components of genetic progress, assisted reproductive techniques are central to modern cattle selection schemes. Of these techniques, ovum pick-up with adjoining in vitro fertilisation (OPU/IVF) has been predominant since 2016 (IETS). While selection of elite cattle is mainly decided by total merit indices composed of production, conformation, and functional traits suitability of donors to assisted reproduction techniques may counterbalance lower genetic merit, donor facilities and OPU/IVF labs costs. The aim of this study was to assess the opportunities offered by selection indices for donors' performance in an OPU/IVF program based on the total number of oocytes collected, viable embryos produced, and development rates up to the blastocyst stage per session. In total, 9,099 OPU sessions performed from 1,596 Holstein donors between April 2018 and August 2023 in a donor station in Northwest Germany were used to calculate genomic parameters using the blupf90 family program and investigate the genetic background of results from assisted reproductive techniques. Heritabilities were highest for oocytes collected ($h^2 = 0.24-0.26$), in the middle range for viable embryos ($h^2 = 0.13-0.14$), and lowest for the development rate ($h^2 = 0.09$). GWAS was performed using postGStf90 and a QTL on BTA11 (92,317,777-97,858,330 bp, ARS-UCD1.3) was identified to explain variance for oocytes collected. Positional candidate genes were aligned to reproductive physiology and verified from former described studies. In summary, selecting donors by their genetic ability to produce embryos using OPU/IVF could improve breeding schemes efficiency in the future.

Effects of moderate exercise regimen on reproductive development of replacement beef heifers reared in drylots at a high stocking density

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This study evaluated growth and reproductive development of beef heifers reared at a high stocking density and provided with access to an exercise area. Angus × Brahman heifers (n = 180) were ranked by age (264 ± 1 day) and body weight (BW; 200 ± 2 kg) on day 0, and assigned to a) 1 of 12 drylot pens (10×14 m pens; 10 heifers/pen) resulting in a stocking density of 14 m²/heifer (DENS), or b) 1 of 6 paddocks (2-ha paddocks; 10 heifers/paddock) resulting in a stocking density of 2,000 m²/heifer (CON). On day 15, 6 DENS pens were randomly selected and given access to an exercise area (DENS-EX) for 1 h three times weekly. Negligible forage was available for CON heifers, whereas all heifers received the same limit-fed corn-based diet. Heifer shrunk BW was recorded after 16 h of feed and water withdrawal on days -3 and 225 for BW gain calculation. Heifers were also fitted on day 0 with a pedometer behind their shoulder and an ear tag to record behavioral responses. Pedometer results were recorded and blood samples were collected weekly. Hair samples were collected from the tail switch every 28 days. Steps per week were greater ($P < 0.05$) for CON vs. DENS and DENS-EX, and greater ($P < 0.05$) for DENS-EX vs. DENS heifers. Heifers from CON spent more ($P < 0.05$) time active, ruminating, and eating vs. DENS and DENS-EX. No treatment effects ($P \geq 0.91$) were detected for heifer BW gain. A treatment × day interaction was detected ($P < 0.01$) for hair cortisol, as CON had less ($P \leq 0.05$) hair cortisol vs. DENS and DENS-EX heifers beginning on day 56. Heifers from DENS experienced delayed puberty attainment (treatment × day; $P < 0.01$) vs. DENS-EX and CON. A greater ($P \leq 0.05$) proportion of CON heifers were pubertal by day 105 vs. DENS and DENS-EX, whereas a greater ($P < 0.05$) proportion of CON and DENS-EX heifers were pubertal by day 154 vs. DENS. Collectively, providing heifers with an exercise area alleviated the negative effects associated with high stocking density on their puberty attainment. Research supported by USDA NIFA AFRI (2021-67015-34083).

Session 41

Theatre 3

Seminal cytokine profiles as markers of bovine sperm quality

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Cytokines orchestrate pathophysiological inflammation and priming of the cervix/uterus for embryo implantation. This study aimed to determine if variations in standard spermatozoa (spz) parameters were reflected in seminal plasma cytokine concentrations. Individual bull semen ejaculates (n=83) were collected on-farm and split; (i) diluted 1:1 in PBS with 0.5% polyvinyl alcohol and protease inhibitor and snap frozen for cytokine analysis, and (ii) profiled for spz concentration, normal morphology, motility parameters, mitochondrial activity and acrosome integrity. Samples from (i) were analysed for interleukin (IL)-1a, IL-1b, IL-4, IL-6, IL-8, IL-10, IL-17a, IL-36a, inflammatory protein (IP)-10, interferon-inducible T cell chemoattractant (I-TAC), macrophage chemoattractant protein (MCP)-1, macrophage inflammatory protein (MIP)-1a, MIP-1b, thymus-expressed chemokine (TECK), tumour necrosis factor (TNF)-a and vascular endothelial growth factor (VEGF)-A by a combination of enzyme-linked immunosorbent assay (IP-10, ITAC, TECK) and fluid-phase multiplex immunoassay (other analytes). All analytes were detectable except IL-4 and I-TAC. Random forest-based analysis with 5-fold cross validation and feature ranking for each objective revealed normal morphology and motility parameters could be identified reliably using only 5 cytokines. Initial data showed positive correlations between a subset of cytokines and spz concentration, motility parameters and acrosome integrity. Statistically significant ($P < 0.05$) negative correlations were noted between acrosome integrity and certain cytokines. These findings suggest that seminal plasma cytokines show potential for use as surrogate indicators for conventional spz parameters and reflect the spz quality of bull ejaculates.

From womb to bloom: conception, early life and maternal effects on boar semen production

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This study aims to quantify conception, early life and maternal effects on boar semen traits. Ejaculates were collected between 2009 and 2022 and evaluated using a Computer-Assisted Sperm Analysis (CASA) system. The phenotypic dataset included records from 449,966 ejaculates collected from 5,692 boars from a synthetic line. Pedigree information included 17,701 animals spanning 26 generations. Conception and early life effects included dam's parity, age of the dam and sire, and litter of origin effects such as gestation length, litter size, litter sex ratio, number of piglets born alive and number of litter mates at weaning. Additionally, length of rearing period and weight gain of the boar during rearing were included. A univariate analysis with a repeatability model accounting for fixed effects at collection, including age of the boar and interval between collections, was used to individually test conception and early life effects. A subset of 230,655 ejaculates from 2,837 boars were used to estimate a maternal environmental effect on semen traits. Conception and early life effects reported suggestive effects on a limited number of semen traits. Significant maternal environment effects were estimated on sperm motility traits, explaining between 2.3 and 5.7% of the phenotypic variance, resulting in lower estimates of heritability. Our findings reveal suggestive effects of early-life conditions and substantial maternal effects on semen quality traits. This insights emphasizes the importance of parental and early-life conditions on semen quality in boars.

Session 41

Theatre 5

Effect of grazing pastures with rich tannin legumes on ovarian activity in suckling anoestrous beef cows

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The length of the postpartum (pp) anoestrous period determines the reproductive performance, and nutrition is one of the main factors affecting it. This study evaluated the ovarian activity in beef pp anoestrous cows grazing two different pastures. Fifty-six Aberdeen Angus×Hereford multiparous cows and their calves (LW 497±6.8 kg; BCS 4.05±0.04 units; Calf LW 60±1,6 kg) were assigned to each of two treatments on Day 22±0.82 pp: (i) native pasture (NP; n=28); (ii) native pastures improved with Lotus subbiflorus (improved pastures, IP, n=28; 3.6g/100 g DM of condensed tannins). BCS and LW was assessed at calving, onset of treatments and every 28 days. Ovarian activity diagnosis (OAD) was performed at Day 57 and 87 pp. OAD was obtained by ultrasonography classifying cows in: Cycling (presence of corpus luteum (CL)); Superficial Anoestrous (SA, maximum follicle diameter (MFD) ≥8mm); Deep Anoestrous (DA, MFD < 7mm). Natural service started at Day 72 pp and lasted 63 days. Treatments were compared using the PROC GLIMMIX of SAS using a binomial distribution. IP cows had greater LW (P<0.05) and BCS (P<0,001) respect to NP cows at Day 52 (545 vs 522 kg, SEM= 7.1 and 4,6 vs 4,2 u, SEM=0.04) and 78 pp (546 vs 523 kg, SEM=7.7 and 4.9 vs 4.5, SEM=0.05). At Day 57 pp more (P< 0.05) IP cows were in SA respect to NP cows (24/28 vs 16/28) but no differences were observed at Day 87 pp since most of cows in both treatments had presence of CL (25/28 and 26/28 for IP and NP, respectively). Although IP cows gained more LW and BCS than NP, cows in both nutritional treatments were above the BCS threshold to achieve adequate reproductive performance.

Relationship between sperm DNA fragmentation, semen quality and serum mineral concentration of bulls
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The present study analyzed the relationship between semen quality parameters (progressive sperm motility, sperm velocity leading), serum concentration of some minerals (selenium and copper), and sperm DNA fragmentation. Semen and serum samples were collected from bulls (n=18) of similar age and breed reared under identical conditions. The semen was collected from the electroejaculation procedure, while blood samples were drawn from the coccygeal vein. Blood samples were sent to the laboratory to determine selenium and copper concentrations, quantified by inductively coupled plasma mass spectrometry. Semen samples were analyzed using the CASA instrument, while sperm DNA fragmentation was evaluated using Sperm-Halamax® KIT. RStudio was used for data management and statistical analysis. Samples with progressive motility above the median (44,45%) were considered optimal. In those samples, a significant (P<0.05) positive correlation was described between semen quality parameters and DNA integrity. These results showed that the decline in semen quality parameters is linked with a lower DNA integrity, which could be translated into a cause of male infertility. Even optimal samples showed a numerically higher content of copper and selenium than the rest, no significant relationship (P>0.05) between the content of these minerals and sperm DNA fragmentation could be demonstrated. Hence, more research is needed.

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Theatre 7

Genomics of boar semen quality traits in U.S. Large White and Duroc breeds

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Semen quality dictates the efficiency and success of artificial insemination, thus impacting the profitability of swine farming. Despite that, semen quality traits are often overlooked in breeding programs. The present study aimed to estimate genomic parameters of semen quality traits in Duroc and Large White boars and to identify candidate genes associated with total number of sperm cells, sperm motility, progressive motility, and normal morphology through a genome-wide association study (GWAS). Data collected from 2013 to 2023 included 524 Duroc and 639 Large White boars genotyped using four SNP arrays with a density range from 48 to 68K. Only common SNP across all panels were retained (n = 27,350). Fixed effects included in the linear model were breed, year, and collection season; age was included as a linear covariate. Boar ID, fitted to a genomic relationship matrix (G), and the residual were the random terms. Heritability ranged from 0.04 (motility) to 0.19 (normal morphology), while repeatability varied from 0.39 (motility) to 0.57 (normal morphology). Strong and positive genomic correlations were estimated between Motility, Progressive Motility, and Normal Morphology (0.76 to 0.96). Total sperm was negatively genomically correlated to motility (-0.06) and progressive motility (-0.39). The GWAS revealed that candidate genes STIM2, ATP10D, ABCA5, and ABCA9 were associated with total sperm, while EBF1, CTNND2, and NAV2 were associated with motility and PARVA to progressive motility. SNP in KCNC1 and ZNF317 were the most significant for normal morphology. Results could be useful in developing breeding subindices for male fertility to maximize profitability and make informed choices in selection nuclei.

Effects of prolactin on serum hormones and related genes of ewes in the estrus cycle

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Prolactin (PRL) has been reported to affect reproductive performance with rhythmic changes. However, the regulation of PRL on hormones and related genes during the estrus cycle remains unclear. This study randomly assigned 62 healthy post-weaning ewes to the control group (C) and the treatment group (T, bromocriptine, PRL inhibition) after estrus synchronization. The samples of blood and ovary were collected from each ewe during the next natural estrus cycle of the proestrus, estrus, metestrus, and diestrus for the detection of follicle count, corpus luteum (CL) count, hormone secretion, and the expression of related genes. The results showed that PRL inhibition had no significant effect on the days of the estrus cycle ($P > 0.05$). T group significantly promoted the number of large follicles and CL ($P < 0.05$), E2, FSH, ER, FSHR ($P < 0.05$), and the sensitivity of FSH during the proestrus period. T group significantly promoted the number of large follicles and CL than that of C group during estrus ($P < 0.05$) and promoted follicle development by significantly increasing StAR and CYP19A1 expression ($P < 0.05$). T group increased the number of CL than that of C group during metestrus ($P < 0.05$) and promoted follicle rupture and ovulation by significantly increasing LHR expression ($P < 0.05$). T group significantly increased the number of large and middle follicles in diestrus ($P < 0.05$). It improved steroid secretion during estrus by increasing P4 levels and PR, 3 β -HSD, and StAR expression ($P < 0.05$) than that of C group. Bcl-2 was reduced and Caspase-3 increased in T group during proestrus. Bcl-2 increased in estrus and Bax in metestrus. In diestrus, Caspase-3 and Bax increased while Bcl-2 decreased in T group. These results suggested that PRL plays an important role in the estrus cycle.

Session 41

Poster 9

Effect of storage time in epididymis at 5°C on motility, viability and morphology of spermatozoa of European red deer (*Cervus elaphus elaphus*)

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The use of stored semen in reproduction may contribute to significant progress in farm breeding of deer. The aim of the study was to assess the motility, viability and morphology of European red deer spermatozoa stored in the epididymis for up to six days at a temperature of 5°C. Spermatozoa were obtained from the cauda epididymis ($n = 11$) post mortem and diluted in Salomon's diluent. Analysis of spermatozoa included assessment of spermatozoa motility (CASA system), viability (SYBR-14/PI staining) and morphology (Giemsa staining). The tests were carried out on the first day (D0) and on the following days of storage (D2, D4, D6). The analysis of the results showed a significant ($p \leq 0.05$) impact of storage time on the analyzed variables. On D2, a significant decrease in motility and integrity of plasma and acrosomal membranes was demonstrated. However, on D4 the values for the percentage of spermatozoa showing movement (TMOT) and viability were still satisfactory and were 68.0 ± 3.7 and 76.1 ± 2.4 , respectively, and the percentage of spermatozoa showing progressive movement was 17.2 ± 1.8 . On D6, there was a decrease ($p \leq 0.05$) in the values of all analyzed variables, and the TMOT and viability values were 31.0 ± 2.4 and 44.7 ± 6.2 , respectively. The obtained results indicate that European red deer spermatozoa can be stored in the epididymis at 5°C for up to four days and used for reproductive purposes. Funded by the Minister of Science under the Regional Initiative of Excellence Program.

Effect of oxidative stress on reproduction of dairy cattle

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The efficiency of cattle reproduction is negatively affected by climate change, causing heat and, consequently, metabolic or oxidative stress. Another aspect that affects cattle fertility is the quality of ovulated oocytes in dairy cows, which is also affected by heat stress or discomfort and the capacity of their metabolism to overcome these conditions. Markers of oxidation and the antioxidant status of the animals can be analyzed from the blood collected from the monitored animals. In laboratory conditions, biochemical parameters of oxidation status (e.g., total oxidant content; lipid peroxidation products) and antioxidant status (total antioxidant content, involvement of antioxidant enzymes) can be determined. Moreover, we focused on production of heat shock proteins 70, a family of conserved ubiquitously expressed proteins that are essential in protecting cells from the adverse effects of physiological stresses, including heat stress. The variability of these indicators is closely correlated with the conception of dairy cows, which thus has a negative impact on the economy of reproduction and breeding of dairy cattle. Monitoring the metabolism level and optimizing the conditions during oocyte collection can be significantly improved, which leads to the efficiency of the whole process. In our study, we present the results of blood analyses for selected parameters from dairy cows at different phases of their reproductive cycle. These phases are reflected in the analyzed parameters and bring us information for streamlining the controlled reproduction of cattle. Supported by NAZV QK22010270

Session 41

Poster 11

The effect of adding penicillin on the quality of pig semen

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Pig farming is one of the livestock businesses carried out by many people in North Sumatra Province. Artificial Insemination (AI) is an artificial mating technique using semen from superior males and can prevent the spread of disease in livestock. In addition, antibiotics are natural substances that have the ability to inhibit or kill microorganisms in semen dilution. One type of antibiotic that can be used in the semen dilution process is penicillin. Some of the advantages of penicillin are that it can inhibit the growth of gram-positive bacteria. This study aimed to determine the effect of penicillin addition in the pig semen dilution process on the quality of pig semen after 24 hours of dilution. The pig used in this study was a male pig over 2 years old, where semen was collected four times as replications. The method used was a Completely Randomized Design (CRD), consisting of 4 treatments with 4 replications, namely P0 (without penicillin in the diluent), P1 (0.3 gr/l penicillin in the diluent), P2 (0.6 gr/l penicillin in the diluent) and P3 (0.9 gr/l penicillin in diluent). The extender used in this study was the non-antibiotic penicillin Beltsville Thawing Solution (BTS). The parameters observed were Motility, Viability, Sperm Abnormalities and pH. The data obtained in this research was then analysed using Analysis of Variance (ANOVA) and continued with HSD test when significant different was observed between the treatments. The results showed that the addition of penicillin was not significantly different ($P > 0.05$) on pH and abnormalities, but was significantly different ($P < 0.05$) on motility, and very significantly different ($P < 0.01$) on viability of sperm. The addition of 0.3 g/l penicillin was the best level in the pig semen dilution process.

Correlated response to divergent selection for litter size variability in rabbits' semen quality

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In this research, sperm production and quality are evaluated in two rabbit lines selected divergently for resilience. Resilience has been measured as litter size variability. The homogeneous line is selected for decreasing litter size variability and the heterogenous lines is selected for increasing litter size variability. The results obtained show that the concentration (P = 83%) and total production (P = 84%) of spermatozoa is higher in the homogeneous line than heterogenous line. The homogeneous line also presents a higher dose potential for artificial insemination (P = 83%). The homogeneous line presents greater movement (P = 94%) and progressivity (P = 92%) . The kinetic parameters are also favourable to the homogeneous line (VCL, P = 90%; VAP, P = 86%; VSL, P = 83%), however, it shows higher values (42.08 vs 38.74%) in parameters associated with circular movements (WOB, P = 93%). It is likely that this difference between lines is not enough to affect the progressivity of the sperm, where the homogeneous line shows better results. In conclusion, the homogeneous line seems to present better sperm production and quality, although it would be necessary to evaluate a greater number of samples distributed over more weeks.

Reproductive tract morphology and symmetry of farmed common eland (*Tragelaphus oryx*) bulls, and their relationship with secondary sexual traits and social rank

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Common eland bulls are popular game ranching animals for tourism, breeding/live sales, trophy hunting, and meat production. Such lucrative profits from many game species like the common eland, have been recently classified as domesticated species in Southern Africa, supporting the intensification of their farming. However, few scientific information is available regarding the selection criteria for breeding bulls. Thus, this research aimed to provide basic information about the reproductive tract morphology, symmetry, and their associations with secondary sexual traits, social rank, and temperament in common eland. Six eland males (2–2.5 years old; 203 ± 20 kg) and their biometric data were utilised and determined before the animals were culled. Reproductive tracts were collected for morphometric and histological evaluations. Relatively low asymmetry in bilateral reproductive tract structures were found. Age, body weight, and social rank correlated the development of some testicular morphologies and possibly be used for the selection of breeding males, but not temperament and horn length, which should be carefully evaluated. Although, this study has provided baseline information from a limited sample size, further studies should incorporate the assessment of testicular hemodynamics, season, and androgen secretion patterns over a wider age range of populations to provide further information for the management practices of breeding common eland bulls.

Effect of food restriction on rabbit ovarian functions

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The aim of this study is to examine the influence of caloric restriction (CR) on basic rabbit ovarian functions. A total of 16 does were used (8 fed ad libitum and 8 subjected to 50% CR). Morphometric analysis of folliculogenesis was carried out by using histomorphometry. Cell viability, proliferation (accumulation of PCNA and cyclin B1, and BrdU-positive cells), apoptosis (accumulation of bax, caspase 3, DNA fragmented cells), release of progesterone and estradiol were evaluated by Trypan blue extrusion test, quantitative immunocytochemistry, BrdU and Cell Death Detection assays, and ELISA. Proteomic analysis was performed by nano HPLC-Chip-MS/MS method. Diameter of follicle, thickness of theca and granulosa, and diameter of antral and preovulatory follicles were higher in CR group than control one. CR group showed lower percentage of viable cells and apoptotic cells, and the release of progesterone and estradiol. CR promoted proliferation, and altered the expression of 25 regulatory proteins. In conclusion, CR affects basic rabbit ovarian functions, and this effect can be mediated by changes in morphometric parameters of antral and preovulatory follicles, ovarian cell proliferation and apoptosis and release of reproductive hormones. Acknowledgements: This study was supported by funds from MICIN/AEI and FEDER, "Una manera de hacer Europa" with project PID2021-123702OB-I00.

Session 41

Poster 15

Camelina sativa L cake as a source of n-3 fatty acids does not affect the reproductive traits of HF heifers

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The beneficial effects of n-3 FAs on metabolic and reproductive performance have been described in ruminants. Although the advantageous impact of n-3 FAs was shown in dairy cows, especially during transition periods, we used the model of nulliparous heifer to avoid possible interaction of the supplement with lactation and NEB effects. The study investigated the effect of Camelina sativa L cake (CSL), a rich source of n-3 FAs, on a panel of variables. This included profile and concentration of FAs, metabolic parameters (glucose, cholesterol, triglycerides, β -hydroxybutyrate), and metabolic hormones (insulin, glucagon, IGF-1). The number and size of ovarian follicles and selected sex hormones (LH, FSH, AMH, PGF2 α) were also analyzed. 16 HF heifers (11-12 months old) divided into control (8) and experimental groups (8) were included. The diet was supplemented with CSL for 48 days (1 kg per day). Diet supplementation with CSL increased the total n-3 FAs and ALA concentrations and reduced the n-6:n-3 ratio. The rise in glucose and cholesterol was observed, but the remaining parameters including sex hormones as well as follicle growth were not affected. To sum up, n-3 FAs may directly affect glucose and cholesterol synthesis. Regarding reproductive variables, our study confirms published results of experiments involving heifers fed fish oil and linseed. Besides, the profile and concentration of bioactive compounds of CSL may also interact with the FA effect. Supported by NCN Poland, grant OPUS21 UMO- 2021/41/B/NZ9/01256

Implementation of an Artificial Intelligence system for morphological evaluation of spermatozoa in routine bull semen analysis

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Implementation of an Artificial Intelligence system for morphological evaluation of spermatozoa in routine bull semen analysis aUrli S., aCorte Pause F., bDreossi T., aStradaioli G. aDepartment of Agricultural, Food, Environmental and Animal Sciences, University of Udine, Via Delle Scienze 206, 33100, Udine, Italy, susy.urli@uniud.it, francesca.cortepause@uniud.it, giuseppe.stradaioli@uniud.it bDepartment of Mathematics, Computer Science and Physics, University of Udine, Via Delle Scienze 206, 33100, Udine, Italy, talissa.dreossi@uniud.it Morphological characteristics of bull spermatozoa are usually evaluated visually using bright field microscopy according to the guidelines proposed by The Society of Theriogenology for the Bull Breeding Soundness Evaluation. However, the evaluation is time consuming and requires experienced personnel to obtain reliable results. Nevertheless, the artificial insemination industry is increasing the usage of genomic selection scheme for young bulls. Hence, there is a growing need for a more standardized technique to analyze semen quality, particularly for the evaluation of spermatozoa abnormalities that affect semen freezing suitability and fertilizing capacity, both important due to the wide use of frozen-thawed semen. Therefore, we are currently working on an Artificial Intelligence (AI) system for automated classification of microscope-acquired images of spermatozoa. We will use neural networks, specifically convolutional neural networks (CNNs) and R-CNN or YOLO networks, which are able to learn and extract relevant features from complex visual data through segmentation of images. This will allow us to classify spermatozoa morphology (normal spermatozoa and both primary and secondary abnormalities). We will employ a dataset consisting of thousands of images, which will be labeled and annotated with bounding boxes to help the segmentation algorithm. If necessary, we will also exploiting augmentation techniques to enhance its size.

Session 41

Poster 17

Combined transcriptome analysis reveals the effect of prolactin inhibition on epididymal function in cashmere goats

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Male animals cannot lactate, but serum concentrations of Prolactin (PRL) are similar to those of females, suggesting that PRL may have an important role in males. However, the molecular mechanisms underlying the influences of PRL on epididymal function remain unclear. We studied the effects of prolactin on the epididymis of rams by feeding the prolactin inhibitor bromocriptine (BCT) to Yanshan cashmere goat rams. Serum hormone levels of the control group (n=10) and BCT group (n=10) were detected by ELISA, epididymal tissues were collected, hematoxylin-eosin (HE) staining to observe histomorphologic changes, and differentially expressed genes were analyzed by RNA-seq. The results showed that at the end of the experiment, the BCT group significantly reduced the hormonal levels of serum PRL, testosterone and estradiol compared to the control group (P<0.05). HE staining revealed no obvious pathological changes in the epididymis in both groups; afterward, we analyzed their measurements with an Independent Samples t-test and found that in the head of the epididymis the BCT group significantly elevated the epididymal epithelial height and the epididymal duct diameter (P<0.05). RNA-seq analysis revealed a total of 358 Differentially expressed genes in both groups. We screened the following pathways by GO and KEGG analysis including steroid binding, GnRH signaling pathway, Estrogen signaling pathway (ESR), cAMP signaling pathway, etc. Fifteen of these genes were selected for verification, and it was found that the qRT-PCR was consistent with the trend of the transcriptome results. In summary, we found that PRL is involved in the reproductive activity of the epididymis by affecting GnRH, ESR, cAMP and other pathways.

Morphometric study of the goat cervix during stages of natural estrus cycle

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The cervical canal plays a crucial role in ewe reproductive biology as it involves multiple reproductive phenomena such as the facilitation of sperm transports, as well as morphological changes during the stages of the estrus cycle. The study of the cervix is paramount, not only for understanding biological processes but also for herd health practice and management. The morphophysiological of the cervix influences numerous reproductive management such as natural mating, artificial insemination as well as pregnancy, and parturition. This study aimed to demonstrate the morphometric data of the domestic goats cervix in different estrus stages, by using CT scan, anatomical appearance measurement, and classification. In this study, the cervical canal was visualized and the 3D pictures were generated to show the appearance of the cervical canal of different cervical grades. Cervical canal grade 1 shows a straight canal with inter-cervical fold space, making a fishbone-like shape. For grade 2, there is a crooked canal around the second or third fold, with a normal straight canal after the crooked, corresponding to the incompleteness of cervical folds at the level of the crooked canal as shown in the longitudinal section. The canal is long, narrow, and crooked, inter-cervical fold cannot be distinguished in grade 3. The mean number of cervical rings was 5, ranging from 3 – 7 rings. The cervix size of the pregnant (11.08 ± 0.7) was significantly smaller than the follicular (14.42 ± 0.66) and the luteal stage (14.23 ± 0.89) ($P < 0.05$). The average canal size was significantly different among stages ($P < 0.05$). Further study on the collagen distribution was determined by Masson Trichrome. The distinct differences among stages were observed in the muscular layer. The percentage of the muscle in the follicular stage is lowest compared to other stages ($p < 0.05$). The percentage of collagen content in the muscular layer was highest in the pregnant cervix. When considering the histology distribution, it was shown that the collagen fibers were slightly detached from each other in the follicular stage compared to the luteal stage.

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Poster 19

Timing of Pregnancy Loss in Lactating Holstein Cows

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The objective was two-fold: i) to quantify the extent and timing of pregnancy loss (PL) in lactating dairy cows, and ii) to determine the days of pregnancy in which PL should be considered stillbirth. A total of 16 commercial dairy farms were selected. Data from 516,473 artificial inseminations (AI) performed on 142,052 cows and heifers were used. Pregnancy was diagnosed with ultrasound at 30 days after AI. Pregnant cows were reevaluated at 70, 120, and 180 days after AI. Cows diagnosed pregnant and subsequently diagnosed open or AI, were considered to have suffered PL. Distribution of PL was determined by logistic regression using the PROC LOGISTIC of SAS. Determination of days of pregnancy in which calving occurred was classified as stillbirth vs PL based on the ROC plot that maximized the Youden's Index J. Overall, 43.1% of cows were diagnosed pregnant at 30 days after AI. The threshold of 260 was selected as days of pregnancy cutoff because it resulted in the highest Youden's Index. Pregnancy loss between 30 and 260 days after AI averaged 13%. The majority ($P < 0.01$) of PL occurred between 30 and 100 days after AI compared to 101 and 260 days after AI (71% vs 29%, respectively). The percentage of cows with twin parturitions was 4.2% and differed ($P < 0.01$) by lactation group (2.1% vs 4.6% vs 5.6% for cows in the first, second, and third or greater calving). In conclusion, delivery of a dead calf after 260 days of pregnancy should be considered a stillbirth. Pregnancy loss had higher incidence between 30-100 days after AI.

The impact of virtual fence on cattle's learning curve over several weeks with multiple borders

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Research on virtual fences has primarily focused on the initial learning phase aiming to test the ability of cattle to respond to audio cues (AC) at a defined GPS border, allowing them to avoid an electrical stimulus (ES) from a collar, return to a virtual pasture. Knowledge of the ability to adapt over several weeks using multiple and changing virtual borders is limited. This study examined how the number of AC and ES varied over 42 days for a group of seven 12-month-old heifers, naïve to virtual and electrical wire fences, introduced to three successive treatments, with different numbers and positions of virtual borders. Treatment 1 (learning phase): 1 border for 1 week, Treatment 2: 2 borders for 2 weeks, and Treatment 3: 4 borders for 3 weeks. The number of AC and ES was collected by the collars. Two heifers were removed on day 2 for reaching the maximum of 15 ES per individual within the first five days. The highest average number per individual and day for AC (33.4 ± 1.9) and ES (9.9 ± 1.3) was recorded on day 1. For the remaining period (calculated for day 2-42) the daily average number per individual for AC and ES was 5.3 ± 0.3 and 0.4 ± 0.1 . Treatment 1 was recorded with a significantly higher number of ES compared to Treatments 2 and 3. There was a significant decrease in the number of AC per day within Treatment 1 ($P < 0.001$) and 2 ($P = 0.013$), but not for 3. Similarly, for the ES, there was a significant decrease in the number per day within Treatment 1 ($P < 0.0001$) but not for 2 and 3. The study concludes that cattle can learn to avoid ES through AC. Moreover, the level of ES per day remains stable over time after the initial learning phase and is not affected by the number or changing of virtual borders.

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Theatre 2

Virtual and electrical fences – perspectives from a survey for cattle farmers in Norway and Sweden

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Virtual fencing (VF), utilizing GPS collars with sensors to create virtual boundaries for livestock, is currently prohibited in Sweden due to animal welfare concerns. However, it has been used in Norway since 2019. The aim was to investigate the perspectives of cattle farmers in Sweden and Norway regarding animal welfare and behaviour with virtual fencing (VF) and electrical fencing (EF). The research comprises two surveys: 1) gathering experiences from Norwegian VF farmers and 2) investigating Swedish farmers' interest in VF adoption and experiences with traditional EF. Preliminary results indicate that most of the Norwegian respondents report that cattle naïve to VF escape 2–5 times and receive 2–4 electrical pulses (EP) during the first week on pasture. During the same period, cattle experienced with VF seldom or never escape and receive 0 or 1 EP. Throughout the grazing season, animals accustomed to VF were estimated to receive a weekly average of 0, 1 or 2–5 EP by 39, 30 and 16 % respectively. No animal injuries were reported on VF pastures. However, 40 % observed abrasions from the collars. On a scale from 0 to 10, the risk of animal injuries while on pasture with VF was rated at 2.3. Perceived animal welfare with VF compared to EF was rated at 8.3. The learning period for cattle to adapt to VF was on average rated 3.7 days. Half of the Swedish respondents estimated that escapes occur “2–5 times” or “more than 5 times” on EF pasture throughout the grazing season regardless of previous pasture experience. 25% responded that they never witness animals receiving an EP from EF after the first week on pasture, whilst 73% occasionally see it. Most of the Swedish respondents had experience of animal injuries on pasture. On a scale from 0-10, the risk of fence-related injuries was rated 3.3. Based on the findings we conclude that, according to farmers, virtual fences efficiently minimize escapes and injuries compared to traditional electrical fences, indicating its potential for improved livestock management and welfare. However, there is a lack of research on cattle learning electrical fencing, though a majority report seeing occasional electrical pulses after the first week of training.

Drone RGB Image-Derived Vegetation Indices to Predict Yield in Humidicola (*Brachiaria humidicola*) Grass
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Forages are a significant component of ruminant nutrition, which relies on proper pasture management. However, improper management results in overgrazing and an overall decline in pasture productivity, causing poor animal performance and welfare. To prevent this, timely and relevant information on forage yield is necessary for the livestock farmer. Traditional ways of measuring forage yield are laborious, time-consuming, and less accurate for wider areas. In precision livestock farming, using drones for this application is gaining popularity because it offers more operational flexibility. Previous research still has limited application because, despite promising results, they used complicated workflows and very expensive equipment. Thus, there is a need for research to develop a workflow that is accurate yet simple, uses cheaper tools, and is computationally efficient. In this research, a DJI Mavic 3 Classic drone with an RGB camera was used to obtain images. Vegetation indices were extracted from these images as predictors to develop a polynomial multiple regression model for the prediction of forage yield. Results showed that fresh and dry matter can be highly predicted from 20-meter altitude with an adjusted R² of 0.71 and 0.66 (R² of 0.77 and 0.73), respectively, by combining linear, quadratic, and cubic vegetation indices. This demonstrates the potential of an alternative simple workflow, cost-effective tools, and statistical method for yield prediction in *Brachiaria humidicola* pastures.

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Theatre 4

Advancing Agricultural Efficiency: Unmanned Aircraft Vehicle in Soil and Pasture Management

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The utilization of Unmanned Aerial Vehicles (UAVs) has become a powerful method for supporting decision-making in pasture management. Multispectral imaging from UAVs could provide quantitative data, through vegetation indices, on the nutritional composition of pasture that may correlate with soil properties and soil health status. Our goal was to predict pasture and soil chemical composition using multispectral images taken from UAV. Data were collected from 11 farms located in Wisconsin, and the model was trained using indices derived from NIR, Red, Green, and Red-Edge bands. Partial Least Squares, Gradient Boosting Machine, and Support Vector Regression models were used and assessed through leave-one-out cross-validation. Additionally, the average of the predicted values was used as an ensemble model. The model predicted plant crude protein with an R² of 0.68 and a root mean square error of prediction (RMSEP) representing 10.9% of the average observed values. It also showed promising results for predicting total digestible nutrients and net energy for lactation in the pasture, with R² values of 0.78 and 0.79, and RMSEP representing 3.0% and 3.2% of the average observed values, respectively. The fiber content in the pasture was predicted with R² values of 0.80 and 0.69 and an RMSEP of 10.2% and 10.8% of the average observed values for Acid Detergent Fiber and Neutral Detergent Fiber, respectively. The model predicted soil pH with R² of 0.52 and an RMSEP representing 6.4% of the average observed values, and soil base saturation with R² of 0.62 and an RMSEP with represents 12.2% of the observed values. Equipped with multispectral cameras, UAVs facilitate the assessment of soil and pasture quality, enabling remote sensing on a large scale for optimal pasture management. Such development is crucial for generating the data required to enhance our understanding of soil-plant-animal interactions in grazing systems.

Can we use open data to track grazing intensity?

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Grazing intensity represents the contribution of grazing to the level of pressure to which vegetation is subjected. Grazing intensity depends on the livestock load per unit of area and time, as well as on the characteristics of the vegetation. Monitoring grazing intensity can be a powerful tool to avoid the potential negative effects of grazing. GNSS tracking can be used to monitor the spatiotemporal distribution of animals on rangelands, but this technology has several drawbacks, e.g. price and connectivity, which can be limiting for some farms. The purpose of this study was to evaluate the feasibility of monitoring grazing intensity using open data sources. Two hundred GNSS collars were fitted to suckler cows grazing in two farms located in Avila, Spain, between 2017 and 2022. Over 7.9 million GNSS positions were recorded and used to compute kernel density estimates per month and pixel (10 x 10 m). The grazing intensity reference values were obtained by standardising kernel values on the basis of the percentage of animals wearing a collar in each farm and study period. Open data sources were used to characterise the spatial and temporal variability of pixels. Digital elevation models were used to compute slope and aspect. Orthophotos were utilised to calculate the distance to water sources and the percentage of grass coverage per pixel. Sentinel-2 imagery was used to analyse the temporal dynamics of grass by computing various vegetation, soil, and water indexes. Pearson correlations were calculated between standardised kernel values and pixel characteristics. Twelve features of pixels were found to have significant correlations with grazing intensity, as estimated from kernel values. However, all correlation coefficients were very low. The monthly average of the Normalized Difference Moisture Index (NDMI) and the Soil Adjusted Vegetation Index (SAVI) had the highest positive correlations with grazing intensity. Conversely, slope and distance to water showed the largest negative correlations. No single pixel feature had a large effect on grazing intensity on that pixel. However, a multivariate approach may help to explain a larger proportion of the variance of this complex phenomenon.

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Theatre 6

Digital ear tags in remote monitoring – exploring new (ab)use cases

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Sensor ear tags (SET) are promising tools for aiding with herd management. Apart from technical challenges, reporting of processed data rather than raw data streams, leads to a thinned or aggregated representation of sensor data. Consequently, available devices excel for specific areas of application. We outline a multi-year effort to successively evaluate SET devices, trial use cases, and test fringe applications. While the long-term goal remains the automation of the mandatory national animal tracing system, we report here on near-term, opportunistic gains based on existing technology for driving sector value. SET were first assessed under different holding conditions for their potential to support automated reporting on cattle. In a second phase, herds of cattle and small ruminants were equipped with commercial SETs on a collar during summering. Analyses were performed on the collected low-frequency data, comprising GPS positions and metrics of activity calculated from accelerometer raw data. The processed data was analysed for its potential to accurately describe herd behaviour and detect anomalies (i.e. predator presence) as this is representative of the data streams a producer purchasing the technology would receive. Herd activity analyses suggest that herd management practices influence daily behaviour pattern, especially level of nocturnal activity. Spatial metrics reflect constraints of available pasture. Interactions of animals with predators are not conclusively resolved in the data. During all test phases, technical and analytical discussions took place between Swiss and Australian stakeholders, representing an industrial user; and the producer – a spin-off – and their original research base institution. Incorporation of dedicated research devices in field trials, and raw data sharing back to the research institution, can contribute to a wider validation of algorithms under hitherto uncovered conditions. A comparative, increasing ease for industry of advancing software and embedded algorithms might allow to translate empirical findings of users into new, dedicated metrics for SETs, to summarise on-device raw data with more problem specificity.

Tracking bull reproductive dominance by measuring male-female interactions using Bluetooth Low Energy
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In cow-calf grazing systems, natural mating is common. Cows are allocated with several bulls, allowing for competition among bulls to mate with cows in heat. Some bulls may exhibit dominance and mate with a large proportion of cows, which can result in a decrease in genetic diversity among calves. Tracking bull reproductive dominance can be time-consuming as it requires visual observations. Paternity tests based on SNPs sequencing can be used, but this may be limiting for some farms. The aim of this study was to assess the effectiveness of proximity sensors based on Bluetooth Low Energy (BLE) in identifying bulls with dominant reproductive behaviour. 53 cows were equipped with BLE beacons placed on a collar, while 3 bulls were equipped with BLE readers programmed to scan and register a maximum of 10 beacons every 30 min. Both cows and bulls were placed in the same paddock allowing natural mating. Cows were observed twice daily to detect signs of heat. Estrotec® patches were used as complementary tool to detect oestrus. Paternity tests were carried out on each bull using 19 SNPs to determine reproductive success. Number of readings (nr) per cow and day for each bull and average time interval between readings (avg gap) were used as indicators of male-female interactions. Significant differences were observed in nr and avg gap on days with cows in heat. Oestrus days exhibited an increase of 9 readings per cow and a decrease in the avg gap by 120 minutes. Regarding dominance behaviour, one of the bulls exhibited clear dominance over the others, siring 42/53 calves born. That bull had the highest nr and the lowest avg gap. This supports the hypothesis that BLE data could be used to monitor dominance in reproductive behaviour. Differences in behavioural indicators between the dominant bull and the others were higher on days with few cows in heat. When multiple cows were in heat simultaneously, there were increased opportunities for subordinate bulls to mate. BLE data may be used to monitor and optimise male-female interactions, reducing or limiting dominance. This could be achieved by separating dominant bulls or favouring oestrus simultaneity.

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Theatre 8

Improving animal health and welfare by using sensor data in herd management and dairy cattle breeding – a joint initiative of ICAR and IDF

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Digitalisation is advancing with rapid developments in farm technologies, which has the potential to revolutionize and to improve the long-term sustainability of dairy production. This has motivated the International Committee of Animal Recording (ICAR) and the International Dairy Federation (IDF) to start a joint initiative aiming at providing guidelines and best practices for using data from sensors across systems and applications, with a focus on functional traits such as health and animal welfare. The key partners are the ICAR Functional Traits Working Group and the IDF Standing Committee of Animal Health and Welfare who have formed a network of representatives from various stakeholder and leading scientists. Research and approaches to improve the usability of data are discussed to promote knowledge transfer and practical implementation in the dairy industry. Experiences and best practices are exchanged, and recommendations for the use of sensor data are being elaborated. The results will be broadly disseminated through ICAR and IDF avenues. Furthermore, the collaborations among multidisciplinary experts are enabling a holistic approach to the current challenges faced by the worldwide dairy industry and will facilitate cutting-edge research and innovation. The initiative will be presented, with a progress report on reference standards, harmonised definitions and terminology, as well as recommendations and best practices regarding data cleaning and editing and definition of novel traits using data from sensor technologies in herd management and genetic evaluations.

TechCare: Technologies to manage the welfare of sheep and goats – from pilots to large-scale studies

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An animal communicates its welfare state through changes in behaviour and physiology. TechCare explores potential technologies to manage the welfare of sheep and goats along the value chain. After stakeholders prioritised welfare issues in the nine project countries, technologies were identified, selected and tested in sheep and goat pilot farms in various environments. Thirteen technologies were evaluated in 5 countries (France, Israel, Italy, Norway, UK). Welfare assessments were also carried out. Five technologies were subsequently retained for further evaluation on commercial farms in 5 countries: indoor/outdoor weather stations and EID tags/readers (France, Greece, Ireland, Romania, Spain); automatic weight crates (Ireland, Spain); milk tank weighers and milk meters (France, Greece, Romania, Spain). Between 10 and 15 farms in each large scale country have been recruited, and welfare assessments are being undertaken, alongside data recorded by the technologies. This will guide the selection of optimal technologies for managing welfare on small ruminant farms.

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Theatre 10

Transferring a 3D imaging POC to beef cattle breeding stakeholders to do on-farm high-throughput phenotyping: the PHENO3D example

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In the French suckler cattle industry, genetic enhancement relies heavily on phenotyping a large animal population. Calves, around weaning, are weighted and their morphology assessed by visual scoring done by advisors affiliated with the Eliance federation. The PHENO3D project endeavors to revolutionize this process through the development of a 3D scanner integrated with Artificial Intelligence for automated phenotyping. This abstract delineates the pivotal roles played by stakeholders in the PHENO3D project: the French Livestock Institute, 3D Ouest, an electronics design department, and two key breeding stakeholders—the national federation of breeding societies, Races de France, and the national federation of performance recording, Eliance. It elucidates on the synergies fostered amongst these entities, facilitating rapid co-development of a tailored 3D scanning solution for breeding stakeholders. Furthermore, it explores how collaborative efforts at both national and local levels have facilitated the collection of a diverse range of phenotypic data crucial for training the AI algorithms responsible for phenotype prediction. Lastly, it underscores the significance of collaborative solution development as pivotal to its future adoption. The PHENO3D project not only spearheads innovation within the French livestock industry but also paves the way for synergistic collaborations across research, technology firms, and breeding stakeholders. It aspires to serve as a model, inspiring similar collaborative endeavors between public and private entities worldwide.

Guideline to validate sensor output of animal-based measurements

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Sensor technology is used in precision livestock farming applications to monitor amongst others animal health and welfare. However, in order to effectively use sensor data for welfare monitoring, the sensors need to provide valid and reliable measurements and the measured data needs to be relevant for animal welfare. Thus, sensor validation is crucial for applications in commercial and research settings. For this validation process, there is a need for a standardized procedure, so that the performance of validation trials are comparable and can lead to a reliable outcome. In the context of the aWISH (HEurope, 101060818) and Clearfarm (H2020, 862919) projects, a guideline is set up to develop a standardized protocol for sensor validation, divided into steps organized into two main blocks. Block 1 is dedicated to output validation, meaning whether the sensor is measuring the output it is designed to measure accurately. Block 2 is dedicated to welfare relevance validation, meaning whether sensor output can provide information regarding animal welfare and can be used as an animal welfare indicator. This presentation will focus mainly on Block 1, which includes different steps: (1) description of the sensor output, (2) determination of the gold standard and reference output, (3) determination of sample size and statistical analysis, (4) limitations of the technology. The guidelines will be illustrated using a number of case studies in cattle, pigs and poultry covering cameras, microphones, wearables, load cells and a combination of sensors.

AgrifoodTEF's pioneering EU service provider from research to industry for the development of Artificial Intelligence and robotic solutions

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The AgrifoodTEF project positions itself at the forefront of innovative synergies in the agricultural sector by promoting the development and validation of cutting-edge solutions based on artificial intelligence (AI) and robotics. By bridging the gap between researchers and private companies, this project marks a significant evolution in applied research, where research institutions offer customized services to industrials to support the development of advanced technologies. A tangible example of this collaboration is seen at Derval experimental farm, where video monitoring solutions for cattle are being assessed. Preliminary discussions between researchers and Ag-Tech companies help to precisely define technical and technological requirements such as the optimal placement of cameras, image quality requirements, and necessary data storage capacities. The design of services offered to companies is then modular, allowing customization according to the specific needs of each company. The services range from providing raw video recordings to external validation of the developed solution with the possibility for the company to add services such as provision of complementary data, video annotation, co-construction of test protocols and more. This modular approach ensures companies to build a service tailored to their objectives. This collaboration between research and industry results in a win-win relationship. For the research sector, it enhances the added-value of experimental facilities and asserts its expertise and experimental know-how. For companies, it offers the opportunity to advance in the development of their solutions and to test them under real conditions through the expertise of recognized researchers. AgrifoodTEF leverages the access to the European network of Testing and Experimental Facilities. AgrifoodTEF emerges as a European network offering a range of services to Ag-Tech companies, highlighting synergies and the diversity of production situations, thereby enriching the agricultural sector with innovative solutions that are tested to be market-ready.

Innovative Orchestration in Sheep Farming-IOSheep 4.0: Ensuring complete welfare, ecofriendly, quality and traceability of lambs

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IOSheep 4.0 marks a significant evolution in conventional sheep farming methods by incorporating cutting-edge technologies like the Internet of Things (IoT), artificial intelligence (AI), and blockchain. This study presents an innovative orchestration model that leverages these technologies to create a holistic and sustainable sheep farming ecosystem. By deploying IoT devices, we monitor the health and welfare of the sheep in real-time, ensuring prompt medical interventions and optimal living conditions. AI algorithms analyze data to predict health issues, optimize feeding strategies, and enhance breeding practices, contributing to improved lamb quality and welfare. Furthermore, blockchain technology offers a transparent and immutable record of the lamb's lifecycle, from birth to processing, ensuring traceability and consumer trust in the quality and safety of the lamb products. Our model also incorporates eco-friendly practices, including sustainable feed sourcing, waste management, and land use, to minimize the environmental impact of sheep farming. The implementation of our innovative orchestration in sheep farming demonstrates significant improvements in lamb welfare, quality, eco-friendliness, and traceability. This approach not only addresses the growing consumer demand for ethically produced and sustainable meat products but also sets a new standard for the sheep farming industry, paving the way for more responsible and sustainable agricultural practices.

The current use and demand for tracking systems on German grazing cattle farms

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Various systems based on triangulation and ultra-high frequency have been in use for several decades for the monitoring of cattle in a barn. In contrast, monitoring of grazing cattle is currently mainly based on the use of GPS and GSM technology. Due to high power consumption for positioning and data transmission, these systems are more suitable for extensive grazing areas. To gain an insight into the current situation, use and requirements of tracking systems on German grazing cattle farms, a nationwide online survey was conducted in 2023. The survey was carried out using LimeSurvey and covered the topics of 'basic farm data', 'cattle and pasture management' and 'use of tracking systems'. Following a plausibility check, a total of 242 farms were included in the analysis, of which 46 % were full-time farms and 54 % were part-time farms, with an average herd size of 114 and 26 cattle, respectively. The most common form of grazing on participating farms was rotational grazing as all-day grazing with a grazing period of 7 months per year. Only on dairy farms half-day grazing was more common (38 %) than full-day grazing (32 %), with the rest performing other grazing times. The most common practice was to carry out a separate control (not combined with feeding and milking times) of the animals on pasture, generally once a day. Only 2.5 % of the farms surveyed used a tracking system for cattle on pasture, but approximately 20 % were interested in using such a system. Most of the cattle tracking systems already in use on the pasture were GPS-GSM tracking systems. High acquisition costs, installation effort and lack of a suitable tracking system for grazing cattle on the market have so far hindered its use in practice.

Veterinarians' perceptions on Precision Livestock Farming application in pig health management

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Data-driven strategies meeting veterinarians' needs can facilitate decision-making in animal health management. This study evaluated veterinarians' goals and their current use and needs of data technology in pig health management. Focus groups were completed in Ireland (n = 8 participants), the Netherlands (n = 7) and Spain (n = 12). Written transcripts of these discussions were analysed with thematic analysis. Veterinarians' goals were to control/prevent disease, being the reduction of antibiotic use, an ongoing challenge. Veterinarians highlighted the potential of data-driven strategies in their daily work. Several structural barriers were however recognized for the current use of data tools, such as lack of convenient access to farm data requiring later transfer to digital softwares. Veterinarians' needs included user-friendly and intuitive data tools to share, manage and analyse data; integrated tools linking different data sources; early warning tools visualizing geographic disease prevalence to support detection and control of diseases; real-time data tools visualising data from farms, slaughterhouses and labs to track pig health status. This study uncovered barriers and opportunities to veterinarians' data use. To facilitate data utilization by transforming raw bytes into useful information, tools for intuitive analysis and visualization with standardized and timely updated and contextual information are in high demand by veterinarians.

Optimizing Low-Frequency Ultrasound for Enhanced Sow Pregnancy Diagnosis Using Vision Transformer-High (ViT-H) Approach

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In the pursuit of advancing swine reproductive management, our study introduces an innovative application of the Vision Transformer-High (ViT-H) model to enhance sow pregnancy diagnosis using low-frequency ultrasound imaging. With the challenges of traditional diagnostic methods, our research leverages deep learning to address the resolution limitations inherent in low-frequency ultrasound devices prevalent in modern livestock farming. Utilizing a dataset of 5,792 non-pregnant and 18,086 pregnant sow ultrasound images across different gestational periods, we applied the ViT-H model under various conditions. The study's breakthrough was the ViT-H model's performance in identifying early to mid-gestation stages with notable accuracy. Particularly, for the crucial early gestational phase of 22-25 days, the ViT-H model exhibited an outstanding accuracy of 87.778%, specificity of 88.754%, and an F1-score of 88.172%, following data augmentation with AutoAugment. These metrics significantly exceed traditional methods, highlighting the potential of transformer-based architectures in veterinary diagnostics. Our findings underscore the transformative impact of integrating AI with veterinary science, presenting a cost-effective, accessible, and accurate solution for early pregnancy detection in sows. This advancement not only propels the efficiency of livestock management but also contributes to the sustainability of pork production.

Predicting the botanical composition of pastures using Near Infrared Spectroscopy

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Livestock diet is crucial for both animal production and health, significantly impacting the quantity and quality of animal-derived products. Grass serves as a primary food source in extensive ruminant farming systems; therefore, the composition of forage is essential for efficient production processes. Near-infrared spectroscopy (NIRS) technology offers a rapid, reliable, and cost-effective method for nutritional analysis without the need for toxic reagents and ensuring complete sample recovery. This study focused on developing a calibration model using NIRS to determine the botanical composition of pastures, specifically the percentage of grasses, legumes, and other weeds. A total of 102 controlled mixture samples from pastures harvested in eight different farms from the Southwest of Spain were created as follows: after fresh harvesting, the plants were manually separated into the three botanic groups (grasses, legumes, and other weeds), and then dried, ground, and sieved. Experimental mixtures were afterward created based on a predetermined percentage table ranging from 0% to 100%, thus resulting in a diverse sample set. NIR spectra and the botanical composition were used to develop Modified Partial Least Squares (MPLS) regression equations, using 90% of the samples to calibrate, and 10% to validate the models. High predictive accuracy models to estimate the percentage of grasses, legumes, and other weeds were obtained, with R² values of 0.99, 0.98, and 0.98, and validation errors of 2.9, 2.8, and 4.2, respectively. Therefore, this study validates the reliability, safety, and efficiency of NIRS technology for pasture analysis and to assess the botanical composition of pastures. This project has received funding from the European Union's LIFE program under Grant Agreement LIFE17 CCA/ES/000035.

Session 43

Theatre 1

Genomic selection in the era of digital phenotyping

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Promoting sustainable breeding programs requires several measures, including genomic selection and continuous data recording. Digital phenotyping can be used to track animal activity and behavior like feeding and walking time and distress continuously. Coupled with machine learning techniques, any feature of interest can be extracted and used as phenotypes in genomic prediction models. It can also help define novel phenotypes that are hard or expensive to measure by humans. For the already recorded traits, it may add extra precision or lower phenotyping costs. One example is lameness in pigs, where digital phenotyping allowed moving from a categorical scoring system to a continuous phenotypic scale, resulting in increased heritability and greater selection potential. Additionally, if an early digital phenotyping behavior is genetically correlated with a trait of interest measured later, selection decisions can be made earlier. One example is the strong, negative genetic correlation (-0.7) between distance traveled and average daily gain in pigs. Conversely, computer vision may add noise to the phenotype for some production or carcass traits, as correlations with the traditional records can be as low as 0.9. In this talk, we will review the benefits and opportunities of digital phenotyping, together with experiences in data analysis and quality control. Finally, we will discuss how to account for the inaccuracy of some phenotypes in genomic prediction models. Overall, digital phenotyping is a promising tool to increase the rates of genetic gain, promote sustainable genomic selection, and lower phenotyping costs.

Comparison of the potential of cow-side blood tests, MIR spectra and wearable sensor devices for breeding for metabolic health in dairy cows

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Metabolic health and in particular ketosis in the early lactation plays an important role in dairy cow management. Due to the low frequency and heritability of clinical cases, breeding for cows with better metabolic health is challenging. Thus, detection of subclinical cases as well as the use of readily available data from milk recordings and automation technologies such as sensor technologies or automatic milking systems may support the development of (auxiliary) traits for metabolic stability in dairy cows. Within the COMET-Project D4Dairy β -hydroxybutyrate (BHB) concentrations from cow-side blood tests measured with the hand-held WellionVet BELUA device at 7 and 14 days postpartum (8,329 test results), mid infrared (MIR) spectra from routine milk recordings up to 30 days postpartum (4,984 records) and data from two different sensor systems (Lely and smaXtec) were made available for 3,717 Fleckvieh dairy cows from 67 farms. Data were collected between October 2019 and January 2022. Sensor data comprised information on rumination and eating time, an activity index, reticular temperature, and number of drink cycles. The following traits were defined for the analysis of genetic parameters: BHB-values and log-transformed BHB, ketosis traits based on BHB-values (1: ≤ 1.2 , 2: 1.3-2.9, 3: ≥ 3 mmol/L and 0: ≤ 1.2 , 1: >1.3 mmol/L, respectively), MIR-predicted ketosis risk from milk recordings (KetoMIR) and mean value and standard deviations of sensor variables on the day of the blood test and milk recording, respectively. Genetic parameters were estimated fitting bivariate models in VCE6. Heritabilities for BHB-value derived traits ranged from 0.1-0.2 and was 0.11 for KetoMIR. Genetic correlations between KetoMIR and BHB-value derived traits ranged from 0.36 to 0.63. Heritabilities for sensor traits ranged from 0.11 to 0.56 and high genetic correlations between activity derived traits and the binary ketosis trait (-0.49 to -0.70) were estimated. Standard errors were low for all estimations.

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Theatre 3

Can daily rumination time be used to breed for resilient Holstein cows?

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Breeding for resilient cows holds the potential to mitigate the impact of external stressors, thereby reducing additional expenditures on labor and antibiotics. Identification of resilience indicators currently becomes feasible through monitoring perturbations in cows' daily rumination time (DRT) and daily milk yield (DMY). The objective of this study, therefore, was to define resilience indicators from DRT in lactating cows, and to investigate the difference in genetics with resilience indicators from DMY. 4,479,754 DMY records of 9,640 cows were used, of which 6,557 cows had in total 1,929,920 DRT records. Resilience indicators were defined based on deviations from a quantile polynomial regression model on DRT and DMY. Indicators were the log-transformed variance (LnVar), the lag-1 autocorrelation (rauto) of daily deviations, the average size of deviations as a ratio to the expected curve value (rper) and the mean of deviations (devper) within the observed perturbations. Genetic parameters for the four indicators were estimated with repeatability animal models. Results showed that the four indicators had a low to moderate heritability (0.09-0.20) and repeatability (0.19-0.40) for DRT, which were comparable in magnitude to indicators derived from DMY (0.10-0.32; 0.22-0.50). The genetic correlations of the same resilience indicators between DRT and DMY ranged from 0.21 to 0.37, indicating they can be used complementary to indicators from DMY to breed for resilient dairy cows.

Use of machine learning methods to predict feed efficiency traits in dairy cows using phenotypic and genomic information

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Predicting direct production values of dairy cows at farm level is difficult. This study focused on the prediction of feed intake (FI), energy balance (EB) and feed efficiency (FE) of Holstein-Friesian cows based on individual phenotypic and genomic information. Different supervised learning algorithms such as Random Forest (RF), Support Vector Machine (SVM) and Extreme Gradient Boosting (GB) were tested. Milk yield, milk fat and protein content, lactation number and live weight between the 100th and 150th day of lactation of 1684 animals as well as a herd-year effect served as phenotypic input information. In addition, for all animals, genotypes of 45,613 SNPs were available. Aiming in reducing model complexity the genomic relationship matrix (G) and the principal component scores of the original marker matrix (UD) were used as genomic information. For each trait and algorithm, three different feature sets (FS) were analysed. FS 1 included only phenotypic data, FS 2 included only genomic data and FS 3 contained both phenotypic and genomic data. For the analyses the data was divided into a training set (80 %) and a testing set (20 %). The training set was used for hyperparameter optimization (5-fold Cross Validation, 10 repetitions) of the algorithms. On the other hand the testing set was used to evaluate prediction performance of the best fitted model from the training on new data. The highest prediction accuracy in the testing set was achieved by the SVM when only the phenotypic data was used. Daily FI, EB and FE were predicted with R² values of 0.60, 0.45 and 0.55, respectively. By adding G and UD, an improvement in the prediction quality could only be achieved with the GB. The consideration of further lactation phases is planned.

Comparison of methods to identify resilience indicator phenotypes using across lactation robot milk yield dynamics in the Nordic Red cows

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Resilience is the ability of the animal to either remain unaffected by disruptions or to quickly return to its normal state before facing a disturbance. Therefore, high resilience to disturbances, fluctuating environmental conditions, and various stressors is becoming more important in dairy farming. However, since resilience cannot be directly measured, indicators can be used to assess resilience. Consequently, identifying phenotypes associated with resilience is a crucial phase in improving the resilience of cows through selective breeding. This study was aimed at testing different methods to identify resilience indicator phenotypes based on the milk yield dynamics over lactation using milking robot data. Data were from 15 dairy cows milked by a DeLaval milking robot at the Natural Resources Institute (Luke) Maaninka research dairy farm in Finland, spanning 2 to 425 days in milk. The perturbed lactation model (PLM) and differential smoothing (DS) methods were used to detect and characterize disturbances in the lactation curves using R software. The methods provided distinct phenotypes including the intensity of the speed of the decrease during the collapse phase and the speed of the increase during the recovery phase by the PLM as well as the magnitude of perturbation by the DS. The results from tested methods showed distinct phenotypes, with both the duration and the magnitude of perturbations differing over the lactation period compared to the idealized lactation curves. A comparison of the methods has been discussed, focusing on numerical stability, repeatability, and computational efficiency. This comparison will be extended to a larger dataset to verify the results. Additionally, the process will include identifying correlated (or redundant) variables to refine the phenotypes.

Genetic correlation between residual feed intake and easily measurable plasma parameters in early-fattening young bulls

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Knowing residual feed intake (RFI) early in life of growing cattle represents an interesting opportunity as it would allow both precision feeding and genetic selection. However, measuring RFI is too expensive. Blood plasma biomarkers represent a cost-effective way to predict RFI and identify feed efficient cattle. Previous works have identified candidate RFI plasma parameters that enable differentiation between extreme phenotypes in young bulls. However, their genetic relationships to RFI need to be clarified. The aim of this study is to estimate the genetic parameters of 13 easily measurable plasma parameters identified as biomarkers of RFI in early-fattening young bulls and assess their genetic correlations with RFI. A total of 639 young bulls were fattened in four experimental farms and were fed ad libitum either grass or corn silage-based diet. Metabolites and plasma enzymes were analyzed during the early fattening stage of animals (303 ± 25 days of age) by spectrophotometric methods performed in an autoanalyzer. Wombat software was used to estimate genetic parameters. The plasma parameters significantly correlated with RFI were alanine aminotransferase (0.89 ± 0.26), aspartate aminotransferase (0.50 ± 0.32), creatinine (-0.46 ± 0.28), triglycerides (-0.44 ± 0.32) and total plasma proteins (-0.41 ± 0.36). Among these five plasma parameters, the heritability ranged from 0.15 to 0.34, while the heritability of RFI was 0.21 (± 0.10). These results pave the way for using blood plasma parameters as proxy to implement genomic evaluation of RFI.

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Theatre 7

Genetic parameters for novel climatic resilience indicators derived from automatically-recorded vaginal temperature in lactating sows under heat stress conditions

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We investigated the feasibility of using automatically-recorded vaginal temperature (TV) data for deriving novel indicators of climatic resilience (CR) for breeding more resilient pigs, especially during lactation when sows are at an increased risk of suffering from heat stress (HS). We derived 15 CR indicators and estimated genetic parameters for all traits, including their genetic relationship with sow reproductive traits. The heritability estimates of the CR traits ranged from 0.00 ± 0.00 (SlopeDe: slope for decreased rate of TV) to 0.29 ± 0.04 (HSUB: sum of TV values below the HS threshold). Moderate to high genetic correlations (0.51 ± 0.05 to 0.99 ± 0.13) and Spearman correlations (0.43 to 1.00) of ranks of genomic estimated breeding values (GEBV) were observed for HS duration (HSD), normalized median multiplied by normalized variance (Nor_medvar), the highest TV value of each measurement day for each individual (MaxTv), and the sum of TV values above (HSUA) and below (HSUB) the HS threshold. These five selected CR indicators were lowly to moderately genetically correlated with shoulder skin surface temperature (range: 0.14 ± 0.01 to 0.48 ± 0.04) and respiration rate (range: 0.08 ± 0.01 to 0.50 ± 0.09). The genetic correlations between these five selected CR indicators and sow reproductive performance indicators, i.e. total number of piglets born alive, total number of piglets born, and number of pigs weaned, ranged from -0.733 to -0.175 , -0.261 to 0.086 , and -0.434 to -0.169 , respectively. The individuals with the highest GEBV values (most climatic sensitive) had higher mean skin surface temperature, respiration rate (RR), panting score (PS), and hair density, but had lower mean body condition scores when compared to the bottom individuals. Most CR indicators evaluated are heritable with substantial additive genetic variance. Additionally, individuals with higher CR are more likely to exhibit better HS-related physiological responses, higher body condition scores, and improved reproductive performance under hot conditions. These findings highlight the potential benefits of genetically selecting more heat tolerant individuals based on the CR indicators.

The relationship between microbial composition and feed efficiency in Iberian pigs

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Incorporating data on microbial composition into selection programmes could improve the prediction accuracy of the breeding values. This study aimed to estimate the percentage of the phenotypic variance explained by the microbial composition for feed efficiency and feeding behaviour traits in Iberian pigs. In addition, the heritability of each genus was estimated to determine which genus is more influenced by the host genetics. Pedigree information and 171 faecal microbial samples (16S rRNA) from Iberian pigs were used to compute different models: (a) the genetic model (GM), (b) the microbial model (MM) and (c) the holobiont model (HM). Heritability estimates (h^2) of feed efficiency and feeding behaviour traits of GM ranged from 0.34 to 0.48, while microbiability (m^2) ranged from 0.15 to 0.26 in the MM. When both effects were combined in the HM, h^2 ranged from 0.25 to 0.38, while the m^2 varied from 0.11 to 0.20. These results suggest that when h^2 and m^2 are estimated separately, their estimates may be inflated. On the other hand, the h^2 estimates of each genus ranged from 0.30 to 0.60 when only the genetic effect was considered. However, when the microbial interactions were included in the model, the h^2 decreased notably (0.13 to 0.27). This suggests that the h^2 estimates of genera may be overestimated by the relationship between microbial species. Therefore, not including the microbial interactions in the model could lead to erroneous conclusions. The results of this study highlight the importance of the gut microbiome on feed efficiency and feeding behaviour traits in Iberian pigs. However, further studies are needed to properly assess the effect of the microbiome on critical traits in Iberian pigs.

Machine learning approaches for classifying the Iberian pig strains based on microbiome

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There is an increasing interest unraveling the factors that shape microbiome composition due to its association with complex phenotypic traits in livestock. Increasing evidence points to the influence of host genetic variation. This interest has coincided with parallel advances in the field of machine learning (ML), providing valuable insights into microbial communities. Thus, the present work aimed to develop ML algorithms utilizing microbiome-based features to classify two genetically distinct Iberian pig strains (EE;Entrepelado and RR;Retinto) and their crosses (ER and RE). The study also compares the most important features used in ML model construction, with microbial taxa identified with classical differential abundance (DA) analysis. Fecal samples were collected from 239 castrated males and sequenced with 16S Illumina MiSeq platform. A total of 37 genera were selected to train nine supervised ML classifiers including ensemble, tree-based, linear, and probabilistic models. Classifiers' performances were assessed using area under the ROC curve (AUROC) on the testing set. Five scenarios were explored with varying classification aims. Models built with exclusively purebred animals achieved the best predictive capacity. The highest mean AUROC was achieved by the Gaussian Naive Bayes (>0.85), confirming microbiome distinctions between strains. Important features generated by permutation importance in ML models were partially aligned with the taxa that showed DA between strains. This study combines ML tools with microbiome-derived data to extract meaningful taxa signatures, potentially characterizing strain differences in Iberian pigs.

In-abattoir 3D measurements of beef carcasses for the prediction of saleable meat yield and novel carcass traits

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Currently, most abattoirs across the United Kingdom (UK) and Europe rely on outdated methods for assessing the value of a carcass (subjective assessment of conformation and fat class). As technology advances, not only are machines capable of objectively predicting the same traits, but also alternative, novel traits. This provides an opportunity for new traits of value to be incorporated into systems that comply with current classification schemes. This study explores the use of 3-dimensional (3D) imaging technology and its ability to extract 3D measurements of beef carcasses to predict the saleable meat yield (SMY) of a carcass. Data were collected from 485 beef carcasses from two commercial abattoirs in the UK. A series of 3D measurements (widths, lengths, and volumes) were extracted from images of beef carcasses, captured using a time-of-flight camera. These measurements were used to predict the SMY (kg), which was calculated following butchery and the weighing of individual primal cuts, fat, and trim from each carcass. Initial stepwise linear regression models show that the 3D measurements are capable of estimating SMY with high accuracy when coupled with other information (cold carcass weight and kill date), ($R^2 = 0.94$). The same variables are also capable of predicting the SMY (kg) of the forequarter, flank, and hindquarter with high accuracy ($R^2 = 0.88, 0.86, \text{ and } 0.81$ respectively). Further analysis will explore the use of machine learning techniques for prediction of SMY and individual primals.

Session 43

Theatre 11

Pixels to feed: Digital phenotyping potential for genetic analysis of feed intake and efficiency in Atlantic salmon

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Improved feed efficiency (FE) is a major goal in salmon production since feed constitutes the largest proportion of production and environmental costs. However, genetic studies on the efficiency of feed conversion to daily weight gain are limited, primarily due to difficulty in phenotyping individual feed intake in aquaculture. This study aims to overcome this limitation by (i) quickly and accurately phenotyping individual feed intake in salmon under commercial conditions; (ii) training deep learning model for efficient object (beads) detection and (iii) analyzing the genetic architecture of FE traits including individual feed intake (FI), average daily weight gain (ADG), Residual feed intake (RFI), and feed conversion ratio (FCR). The study was conducted on approximately 700 Atlantic salmon smolts belonging to 35 fish families of the 2017-year class at MOWI Genetics, Norway. The snapshots of individual FI were recorded using X-ray imaging. A deep learning model (YoloV5) for bead detection was trained to estimate the FI accurately and efficiently and then deployed on all X-ray images. The model performed exceptionally well with an R^2 of 0.99, low RMSE and a slope of ~ 1 across training, validation and test sets. Heritability estimates were intermediate to high for FI ($h^2 = 0.20 \pm 0.05 - 0.50 \pm 0.06$) and ADG ($h^2 = 0.47 \pm 0.07 - 0.54 \pm 0.06$), and low to moderate for FCR ($h^2 = 0.08 \pm 0.04 - 0.23 \pm 0.06$) and RFI ($h^2 = 0.10 \pm 0.05 - 0.17 \pm 0.11$), indicating their potential to be improved through selective breeding. We are currently diving into GWAS and genomic prediction to better understand FE genomics.

Genotype-by-environment interaction with high-dimensional environmental data: an example in pigs
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This study investigated using environmental covariates to model correlated herd effects and genotype-by-environment interaction (GE), as well as their impact on the prediction accuracy of genomic evaluations in pigs. We used air, dew/frost, wet-bulb, and earth-skin temperatures, relative humidity, rainfall, wind speed and direction. Average daily gain (ADG) and backfat thickness (BFT) from a terminal pig line were analyzed by 6 different models: MG, which included herd as an uncorrelated random effect; ME30, which considered 30 days of daily weather information to correlate herd effects; and ME100, which used 100 days of daily climate covariates; MGE30, same as ME30 but also considering the GE as the Hadamard product of the genomic relationship matrix and the environmental covariates matrix; MGE100, same as ME100 but including the GE; and MTM, a traditional multiple-trait model with each herd as a different trait. All animals were genotyped and phenotyped; therefore, the statistical model of choice was GBLUP. Validation was based on LR method, and focal animals were those born in the last year. Accuracy, bias, and dispersion did not change among models, except for MTM. Comparing all models against MTM, the accuracy increased by 38% for BFT, whereas only slightly increased for ADG. Among all 11 herds, only 4 showed a genetic correlation stronger than 0.80 for BFT, while this number was much bigger for ADG. When the model has a variable accounting for environmental effects like herd in the case of MG, using weather information to correlate environments or adding the GE term has little benefit for genomic predictions.

Session 43

Poster 13

Association analysis of daily rumination time with hindgut microbiota and metabolites in dairy cows

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The maintenance of rumination behavior is essential for dairy cattle's health and production performance. The gastrointestinal microbiota and their metabolic process may be the key factors shaping cow rumination behavior. However, the characteristics of hindgut microbiota and metabolites associated with rumination time are still largely unknown. Therefore, this study aimed to comprehensively identify the association between cow rumination behavior, hindgut microorganisms, and metabolite. The daily ruminating time measured by Hi-Tag necklet, hindgut microorganisms analyzed by 16S rRNA sequencing, and hindgut metabolites detected by LC-MS of 100 healthy first-parity Holstein cows with the same feeding condition and similar lactation days were available for this study. A total of 68 differential microorganism markers and 177 differential metabolites were identified using the LEfSe analysis (LDA>1, P<0.05) between high (547 min/d) and low (496 min/d) rumination time groups. The correlation analyses between differentiated microbiota and metabolite showed that *Romboutsia* was positively correlated with Morph, Isophorone, 3-Hydroxypicolinic acid, and Quinolinic Acid, and negatively correlated with Diethanolamine; *Howardella* was positively correlated with 5-Acetylamino-6-amino-3-methyluracil, and negatively correlated with Gemcitabine, Heliotron, and Artonin T; *Incertae Sedis* was positively correlated with Diethanolamine; and *Negativibacillus* was negatively correlated with Artonin T. These results increase the understanding of the microbial and metabolic mechanisms of rumination behavior and might help optimize feeding management and selection programs in dairy cattle.

Sensitive pathogens detection using nanozyme-based catalysis amplification on Ag-PSi SERS scaffold

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Food safety and the prevention of foodborne illnesses depend significantly on the quick and accurate identification of pathogenic microorganisms. A bifunctional Pt@Au core-shell nanozyme was designed for the rapid identification of two major pathogens (*Escherichia coli* and *Staphylococcus aureus*) based on a label-free surface-enhanced Raman scattering (SERS) approach. Herein, we utilize their capacity to imitate natural enzymes to oxidize 3,3',5,5'-tetramethylbenzidine (TMB) into an oxidized product (oxTMB), a highly active Raman-reporter-molecule. The number of signal molecules may be considerably increased by catalyzing the conversion of Raman-inactive molecules into reporters using the ultrathin Pt shell. Additionally, the Au core and silver-decorated porous silicon (Ag-PSi) act as an active SERS substrate to boost reporter molecules' signals, prominently enhancing the detection sensitivity. The bifunctional Pt@Au nanozyme coupled to SERS immunoassay achieved low detection limits of 4 and 5 CFU mL⁻¹ for *Escherichia coli* and *Staphylococcus aureus* in an overall 60 min assay duration. Furthermore, the signal amplification, inherent physical features, platform's selectivity and overall shelf-life were thoroughly evaluated while depicting satisfactory performances. Finally, the practicality of the developed SERS bioassay was elucidated in various products with different pathogens concentrations that revealed recovery values of 90-115% with respect to conventional practices. The successful validation of actual samples' analysis emphasizes the platform's reliability, robustness and suitability for practical use, including on-site or pen-side operation.

Usability of raw phenotypic data in selective breeding of dairy cattle on health traits in Slovakia

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The objective of the study was to estimate genetic parameters and breeding values for health traits and somatic cell score (SCS) on the 6 selected farms belonging to the Top 50 in Slovakia. Data for the analysis were collected between the years 2017 to 2022, with a dataset including 8776, 7923, and 7745 cows for the analysis of mastitis, claw diseases, and SCC. For estimation, single-trait animal models were used; considered fixed effects of parity, year, season, herd, breed (single or as interaction), the permanent environmental effect, and the additive effect of the animal. Estimated heritabilities for health traits were low: 0.0209 for mastitis (binary coded 0 healthy/1 ill), 0.0205 mastitis (number of cases 1/6), 0.1550 for SCS and 0.0161 for claw disease (non-infectious binary coded 0/1), 0.0592 (total binary coded 0/1) and 0.0908 (infectious binary coded 0/1). Reliabilities of breeding values were found 0.0665 for mastitis (binary coded and number of cases), 0.1342 for SCS, 0.0335 for non-infectious claw diseases, 0.0341 for infectious claw diseases and 0.0461 for claw diseases total. Significant correlation was observed between estimated breeding values (EBV) for mastitis (binary coded) and mastitis (number of cases) 0.909. Significant correlations of EBV for SCS and mastitis, ranging from 0.290 to 0.357, and among different groups of EBV for claw diseases from 0.0068 to 0.893 were found. Broader involvement of farmers and breeders in data collection and higher uniformity of health trait evidence could lead to decrease of variability, increase of reliability and breeding advancement. This research was funded by DS-FR-22-0016 and APVV-20-0161.

Machine learning and parametric methods for genomic prediction of feed efficiency-related traits

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Genomic selection (GS) offers a promising opportunity for selecting animals for feed efficiency (FE) traits in beef cattle, impacting profitability and environmental sustainability. Here, we compared the prediction accuracy of multi-layer neural network (MLNN) and support vector regression (SVR) against the parametric methods single-trait (STGBLUP) and multi-trait GBLUP (MTGBLUP) and Bayesian (BayesA, BayesB, BayesC, BRR, and BLasso) for FE-related traits. FE-related traits, average daily gain (ADG), dry matter intake (DMI), FE, and residual feed intake (RFI) were measured in 1,156 Nellore animals from an experimental breeding program and genotyped with ~300K markers after quality control. Prediction accuracy (Acc) was evaluated using a forward validation, splitting the dataset based on birth year and using the adjusted phenotypes for fixed effects as pseudo-phenotypes. The MLNN and SVR approaches were trained, splitting the training population in 5-fold. The results show that MLNN, SVR, and MTGBLUP outperformed STGBLUP and Bayesian regression, increasing the Acc by approximately 8.9% with MLNN, 14.6% with SVR, and 13.7% with MTGBLUP. The Acc for SVR and MTGBLUP were slightly different, ranging from 0.62 to 0.69 and 0.62 to 0.68, respectively, and empirically unbiased for both models (0.97 and 1.09). Our results indicated that SVR and MTGBLUP approaches were more accurate in predicting FE-related traits than Bayesian regression and STGBLUP and seemed competitive for GS of complex phenotypes with various degrees of inheritance. Acknowledgment: FAPESP 2009/16118-5; 2016/24228-9, 2017/13411-0 and 2017/10630-2

Session 43

Poster 17

Prediction of blood β -hydroxybutyrate and hyperketonemia based on milk mid-infrared spectra in dairy cows

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Hyperketonemia in early-lactation dairy cows is mainly detected through analysis of associated blood metabolites, such as β -hydroxybutyrate (BHB). Mid-infrared (MIR) spectroscopy could however be a potential alternative to predict blood BHB content and to identify cows at risk for hyperketonemia. Thus, the objective of the present study was to predict blood BHB content in early-lactation dairy cows using milk MIR spectroscopy. The calibration dataset comprised 1005 observations of BHB concentration measured by a handheld device from capillary cow blood on first and second test-day between September 2020 and March 2021. Prediction models were developed based on 212 selected first derivative MIR spectra and test-day variables with BHB content or hyperketonemia class, based on BHB threshold, as dependent variables. Regression and discrimination analyses were conducted applying various machine learning algorithms. An additional dataset was used for external validation. Models developed for quantitative prediction of BHB achieved coefficients of determination ranging from 0.19 to 0.33 in external validation. Discriminant models for hyperketonemia obtained good classification accuracies (sensitivity 0.67-0.71, specificity 0.70-0.73). Results indicate a potential to monitor cows with hyperketonemia using MIR spectral data and to generate novel phenotypes for animal breeding on a large scale.

Dissecting production traits using metabolomics in pigs

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Italian heavy pigs are mainly raised to produce Protected Designation of Origin (PDO) dry-cured hams. To breed pigs with the characteristics of the meat and legs needed for this purpose, specific selection programmes have been developed, using traits related to the overall fat/lean meat content of the animals, to growth and efficiency of the animals and to ham quality (e.g. intermuscular fat and weight loss at first salting). Metabolites represent simple molecular phenotypes that can be used to deconstruct complex phenotypes in their single biological components. We analysed targeted and untargeted metabolomic profiles (up to 1000 molecules) from plasma of Italian Large White and Italian Duroc pigs for which production traits and their estimated breeding values were available. We studied the relationship between all these simple and complex traits via correlation analyses and multivariate modelling. In general, metabolite abundances poorly correlated genetically with production traits (up to $|r_G| < 0.15$), except for a few metabolites (up to $|r_G| \sim 0.4-0.5$). Overall, we obtained a first picture of the correlation between molecular phenotypes and economically relevant traits that could be useful to further exploit their usefulness in designing selection programmes in Italian heavy pig breeds. Acknowledgements: This study has received funding from the European Union's Horizon Europe research and innovation programme under the grant agreement No. 01059609 (Re-Livestock project).

Longitudinal feed intake deviations at the pen level as a resilience indicator for pigs

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Recently, new resilience traits based on longitudinal deviations in pigs' weight, feed intake (FI) and feeding behaviour were developed. However, they require individual repeated observations which is often challenging or costly to record. Therefore, we evaluated longitudinal FI records at the pen level of related pigs as a potential resilience indicator trait. We studied 704 pens during the finishing phase composed of a mixture of 7 to 15 halfsib and fullsib pigs with known pedigree (352 Piétrain sires; 679 hybrid dams). Cumulative FI was recorded weekly at the pen level and studied for pigs of 90-160 days of age. All data were recorded at the same farm between 2020-2023. Longitudinal deviations of FI at the pen level were estimated as the natural logarithm of the mean squared error after linear regression of FI versus mean age of pigs (lnMSEFI). Genetic parameters were estimated using *remlf90* software. The heritability of lnMSEFI was estimated at 11.8%, while the common environment (herd year season) explained 44.5% of total variance. lnMSEFI was genetically correlated with FI ($r_G=0.59$) and daily gain ($r_G=0.46$), not with feed conversion ratio ($r_G=0.00$), and negatively correlated with meat percentage ($r_G=-0.42$). Breeding values for lnMSEFI were significantly correlated with breeding values for mortality ($r=0.23$). Our findings show that longitudinal deviations in FI are heritable, and associated with mortality, showing the potential of lnMSEFI recorded at the pen level as a resilience indicator.

Genetic analysis of milk FTIR spectra in Italian Simmental Cattle

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Fourier Transform Infrared spectra of cattle milk are currently used for investigating and predicting novel phenotypes (e.g., milk fatty acid profile, methane emissions). Due to the huge amount of information contained, the milk FTIR spectrum can be directly considered as a novel phenotype. The present work aims at assessing the genetic component of milk FTIR spectra in cattle. Data were TD records of milk FTIR spectra recorded on 39,271 Italian Simmental cattle. Variance component estimation was carried out with a BLUP animal model that included the fixed effects of herd-test date combinations and parity, and the random additive effect of the animal. A total of 103,878 animals were considered in the pedigree-based relationship matrix. The analysis was carried out on both 1,060 wavenumbers and principal component (PC) scores extracted from original data. In particular, the first four PC were able to explain 76% of the original variance. Heritability estimates for wavenumbers ranged from 0 to 0.19; whereas heritabilities were 0.07 ± 0.01 , 0.05 ± 0.01 , 0.24 ± 0.02 , 0.06 ± 0.01 for PC1, PC2, PC3, and PC4, respectively. These results underline a moderate to low genetic component for milk FTIR spectra variability and confirm that they can be modified by selection.

Session 43

Poster 21

Prediction of nutritional content of black soldier fly (BSF) larvae using multispectral imaging

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The economic viability of BSF production for livestock and pet nutrition heavily relies on the nutritional composition of its larvae, notably protein and fat content, influenced by diet, harvest age, and genetics. Maintaining consistent product quality necessitates continuous monitoring across larvae developmental stages. Selective breeding also offers potential for enhancing BSF larvae's nutritional profile, requiring phenotypic data from potential live candidates. Conventional chemometric methods, while accurate, are costly, time-consuming, and destructive, hindering their suitability for live larvae phenotyping. Hence, there's a pressing need for rapid, non-destructive, and cost-effective techniques to evaluate the nutritional content at different stages of the BSF larvae. Recently, spectral imaging has emerged as a powerful tool for food quality assessment, capturing both spectral and spatial data simultaneously. This study explores the potential of multispectral imaging in predicting individual BSF larvae's nutritional content. With 71 larvae sampled and imaged, prediction models, employing 19 wave bands as features via PLS, exhibited promising predictive performance for lipid mass and lipid percent. With reasonable correlation between measured and predicted parameters for a test set of 21 larvae, the study demonstrates the promise of multispectral imaging in predicting individual larvae's nutritional content, offering avenues for future research in nutritional content prediction from live samples.

Comprehensive analysis of whole-transcriptome and metabolome provides insights into estrus expression in Holstein cattle

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The occurrence and the intensity of estrous expression improved the pregnancy of dairy cows, maximizing economic benefits in farms. Thus, the study aimed to investigate the underlying regulatory mechanism of estrus expression through metabolome and transcriptome integrated analysis. The intensity of estrus expression was assessed based on heat indicators from calving to first estrus (HIC) and interval from calving to first heat (ICH) by neck collar. Nine cows were categorized into three groups: LH_S (low HIC and short ICH), LH_L (low HIC and long ICH), and HH_L (high HIC and long ICH). A total of 8,727 mRNAs, 6,237 lncRNAs, 16,749 circRNAs, and 2,408 miRNAs were quantified and normalized. Results showed 1,266 DERNAs were regulated by a consistent trend, with 764 downregulated DERNAs and 411 upregulated DERNAs in both LH_S and HH_L compared to the LH_L. Furthermore, ceRNA networks were established, revealing bta-miR-671 with the highest number of target genes involved in pathways such as retinoic acid receptor signaling, GnRH signaling, and the estrous cycle. Five overlapped DMs associated with initial estrous events were observed in three groups. Moreover, weighted genes co-expression network analysis (WGCNA) identified three co-expression modules related to the overlapped DMs. The metabolite-RNA correlation network revealed 18 positive correlations (0.67 ~ 0.82, P<0.05) and 138 negative correlations (-0.67 ~ -0.90, P<0.05) between DMs and DERNAs belonging to the three key modules. Overall, this study found distinct regulatory mechanisms underlying the intensity of estrus expression by integrating omics data, offering valuable insights for enhanced fertility traits in Holstein cattle breeding programs.

Unraveling metabolic stress response in dairy cows: genetic control of plasma biomarkers throughout lactation and the transition period

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Breeding cows able to effectively respond to stress could be a strategy to improve livestock resilience and welfare. In this work, the concentrations of 29 plasma biomarkers were used as endophenotypes for metabolic stress response in multiple GWAS approaches on 739 healthy lactating Holstein cows and 88,271 variants. Significant genetic associations were found for plasma γ -glutamyl transferase (GGT) concentration on BTA17, paraoxonase on BTA4, and alkaline phosphatase and zinc on BTA2. On these chromosomes, single-SNP and gene-based chromosome-wide association study confirmed GWAS findings. The signals identified for paraoxonase, GGT and alkaline phosphatase were in proximity of the genes coding for them. Plasma biomarkers concentration largely vary during the metabolic stress of transition period, with an inter-individual variability in the rate of change and recovery time. Genetics may account in part for these differences. To assess this, on a subset of 139 periparturient cows homozygous at three SNPs known to be respectively associated with plasma concentration of ceruloplasmin, paraoxonase and GGT, we compared the plasma immune-metabolic profile at -7, +5 and +30 days relative to calving between groups of opposite homozygotes. A significant genotype effect was found on paraoxonase and GGT concentration at all the three time points. No evidence for genotype effect was detected for ceruloplasmin. Understanding the genetic control underlying metabolic stress response may suggest new approaches to foster resilience in dairy cows.

Phenotyping tools and data collection for the agroecological transition

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Livestock phenotyping has made significant progress in recent years through the increasing use of digital technology. When used appropriately, phenotyping tools can be an important lever in the agroecological transition of livestock production. The GIS Avenir Elevages undertook a multi-species review of recent advances in animal phenotyping and the contribution these new tools could make to the agroecological transition. In particular, we look at the measurement of new traits related to environmental impact (production of greenhouse gases), the efficient use of feed and water resources (intake of concentrates or forages, water, digestibility, etc.), animal behaviour related to welfare (movement sensors or image or sound analysis using artificial intelligence tools, etc.) and health (in particular through "omics" tools). The prospects of continuous measurement of more standard phenotypes (weight, milk or egg production) are also mentioned. The strengths and weaknesses of the different phenotyping technologies and tools will be discussed. The review will 1) provide a multi-species inventory of phenotyping tools for traits, and 2) identify gaps in certain areas and make proposals for research and development actions to fill them.

Session 43

Poster 25

Blood metabolic biomarkers predicted from milk spectra are heritable in Holstein transition cows

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Blood metabolic parameters provide useful information about cows' metabolic status, especially in early lactation. Determining these traits is however costly and requires invasive blood samplings. We used milk mid-infrared spectroscopy to predict blood concentration of β -hydroxybutyrate (BHB), non-esterified fatty acids (NEFA), urea, cholesterol (CHO) and glucose (GLU). Blood and milk samples were collected from 680 cows between 5 and 35 days in milk in 34 multi-breed Italian farms during the morning milking. Partial least square regression was performed for prediction models development and coefficients of determination in cross-validation were 0.64 for BHB, 0.64 for NEFA, 0.85 for urea, 0.53 for CHO, and 0.33 for GLU. Subsequently, we estimated heritability (h^2) with a linear animal model using blood traits obtained from a prediction set (8,277 early-lactation Italian Holstein cows, 374 herds). BHB exhibited the highest h^2 (0.13 ± 0.03), while NEFA the lowest (0.03 ± 0.01). The h^2 of the remaining traits was low too, ranging from 0.04 to 0.08. The minimum and the maximum coefficient of additive genetic variation were 3.1% (GLU) and 20.8% (BHB). Although the prediction accuracy and precision do not allow for punctual determination, predicted blood traits can be still used for selection purposes to guide genetic progress towards a reduced incidence of metabolic diseases.

Evaluating the Efficiency of Phenomic Selection in Dairy Cattle

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Phenomic selection is a relatively new, high-throughput method that uses spectra information from biological samples for phenotype prediction and selection. This study proposes a large-scale application of phenomic selection in dairy cattle using mid-infrared (MIR) spectra, routinely produced from milk samples, to predict milk composition. The objective is to compare the accuracy of the predictions obtained from phenomic evaluation (hyperspectral BLUP, or HBLUP) with those obtained with genomic evaluation (GBLUP). 36,986 milk MIR spectra records were available from three French dairy cattle breeds: Holstein (n = 2,330), Montbéliarde (n = 1,726), and Normande (n = 2,805), to predict eleven traits related to milk production (milk, protein, and fat yields, protein and fat contents), udder health (somatic cell count and clinical mastitis), fertility (heifer and cow conception rates, and calving to artificial insemination interval), and height at sacrum. All traits were evaluated using their yield deviations. The estimated heritabilities of absorbances along the wavelengths of the MIR spectra varied from 0 to 0.5, indicating the potential of using MIR spectra for genetic evaluation. Although the accuracy of the HBLUP predictions is not as good as that obtained with a GBLUP, on average, the HBLUP achieved up to 89% of the GBLUP prediction accuracy for functional traits, 73% for production traits and 42% for type traits. This result suggests that phenomic selection can be a low-cost alternative to the use of genomic information for breeding value estimation in dairy cattle. The next ongoing step of our study is to combine genomic and MIR spectra information to predict breeding values.

Session 43

Poster 27

Genomic evaluation of SA Holsteins incorporating local and foreign genetic markers under two production systems

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Evidence for genotype by environment (GxE) interaction between total mixed ration and pasture feeding systems (FS) for persistency of production has been shown in South African (SA) Holsteins. Analyzing test-day (TD) records from the two FS as separate traits, estimated breeding values (EBVs) from best linear unbiased predictions (BLUP) showed significant re-ranking of sires between FS. The addition of genomic information (single nucleotide polymorphism markers; SNPs) to existing pedigree information to get more accurate genetic relationships amongst animals over the FS may enhance prediction accuracy. The same 3-lactations, TD, random regression model utilized in the GxE study was used to analyze a comparable dataset. The model estimates the (additive) genetic effect of lactation as the function of an average-production (constant) and -persistency (slope) effect over days in milk. First, about 50K SNPs were used to form the genomic relationship (GR) matrix of the local SA Holsteins. Then SNPs from related foreign sires were added in an effort to form a more complete GR matrix. The single-step (SS) BLUP approach was used to form the H-inverse matrix, combining the pedigree and GR matrices, together with TD records from both FS and deregressed proofs of foreign sires to calculate SSBLUP EBVs. These SS-EBVs, their reliabilities, their rankings as well as the genetic parameters over the FS were compared to pedigree-based BLUP EBVs.

Evaluation of muscle transcriptome of beef cattle after intramuscular application of vitamin A
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This study assessed the skeletal muscle transcriptome of calves post intramuscular vitamin A administration at birth. Forty male crossbred calves were studied from birth to feedlot finishing. At birth, 20 calves received a single intramuscular injection of 300,000 IU of vitamin A, while the remaining 20 calves were administered a placebo (control). After weaning at 210 days, all underwent 180 days of feedlot finishing. Longissimus muscle samples were collected for RNA-seq analysis (hot carcass) and meat quality evaluation (48h post-mortem). Total RNA was extracted from 100 mg LT muscle using TRIzol®, analyzed on Bioanalyzer 2100®, and sequenced using TruSeq RNA Sample Preparation Kit (Illumina, USA) on Illumina NextSeq550®. Transcriptome analysis identified 165 differentially expressed genes (DEGs), 59 upregulated in vitamin A group ($P < 0.01$; \log_2 FC [0.5], FDR 5%). Functional analyses revealed enrichment of defense mechanisms, intracellular signaling, fatty acid metabolism, and oxidative processes. Vitamin A led to >50% increase in intramuscular fat ($P < 0.05$) compared to control ($4.10 \pm 0.35\%$ vs. $2.57 \pm 0.27\%$). A trend ($P = 0.07$) towards meat tenderness was noted in vitamin A group (WBSF = 3.92 ± 0.16 kg vs. 4.23 ± 0.15) after 7 days of aging. Vitamin A upregulated key biological processes of transcriptome associated with energy metabolism, promoting IMF accumulation during finishing, which may help to improve meat tenderness and juiciness.

Session 44

Theatre 1

Deciphering the Genomic Architecture of Intramuscular and Subcutaneous Fat Composition in Iberian Pigs
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The genetic architecture of intramuscular (IMF) and subcutaneous fat (SCF) composition in Iberian pigs was investigated to elucidate key factors influencing meat quality traits. Fatty acid profiles play a crucial role in meat quality, affecting fat firmness, intramuscular fat content, and oxidative stability during meat processing. In this study, 703 animals were analysed for fatty acid profiles in subcutaneous fat, 633 of them were also analysed for fatty acid profiles in intramuscular fat. After a CLR transformation for compositional data, we estimated the heritability and the genetic correlations between IMF and SCF for all analysed fatty acids and SFA, PUFA and MUFA. Moreover, we identified the genomic regions associated with largest additive genetic variance for each one of them. Heritability estimates were higher in SCF than in IMF, with varying levels across fatty acids. Genetic correlations between SCF and IMF fatty acids were predominantly positive, except for C20:3 (n-6), indicating a negligible relationship due to its residual percentage. Moreover, multi-trait GWAS identified significant regions associated with fatty acid composition, including genes like FAF2, LIPA, LIPM, and ANKRD1, known for their roles in lipid metabolism. This study enhances our understanding of Iberian pig genetics and provides insights for targeted breeding programs to improve meat quality.

Single marker and haplotype-based GWAS for meat quality and carcass traits in Italian heavy pigs

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The selection and breeding programs of Italian heavy pig breeds have as their main objective the improvement of meat quality and carcass traits useful to meet the needs of the protected designation of origin dry-cured ham industry. The aim of this study was to investigate the association between genetic variants and several meat and carcass traits (e.g., pH, color, water holding capacity, intermuscular and intramuscular fat content, the weight of carcass cuts, back fat thickness, lean meat content, and carcass grading parameters) in a total of ~1300 pigs of the Italian Large White and Italian Duroc breeds genotyped with an SNP array. After assessing phenotypic correlations, single marker and haplotype-based genome-wide association studies (GWAS) were performed. Several genomic regions associated with the investigated parameters were identified, pointing out a few candidate genes. The results will be used to better understand the genetic components of relevant traits useful to address the selection programs of Italian heavy pigs. Acknowledgments: Funded by the Programma di Sviluppo Rurale Nazionale (PSRN) SUIS-2, co-funded by the European Agricultural Fund for Rural Development of the European Union and by the MASAF; and by the European Union – NextGenerationEU under the National Recovery and Resilience Plan (PNRR) – HamCapture, proposal code 202238NP9N – CUP J53D23009570001.

A promising genetic background of pork hemorrhages occurring at slaughter

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Reportedly, the use of inert gases like nitrogen or argon for stunning as well as electrical stunning induces more hemorrhages in the valuable carcass parts than CO₂. In the project for Testing Inert Gases in order to Establish Replacements for high concentration CO₂ stunning for pigs at the time of slaughter (TIGER) these findings could not be confirmed when the animals were stunned with argon. Out of 214 crossbreed pigs, 51 showed hemorrhages in the ham, where their incidence in the CO₂ and argon group was nearly equal, respectively. To investigate a genetic background that makes animals more susceptible to the bleedings, the pigs were genotyped using the GGP Porcine 50 K Array Chip. The data was filtered for 95 % genotyping rate per individual, minor allele frequency > 0.05 and 90 % SNP genotyping rate. The used model for heritability (h²) estimation and GWAS for the occurrence of blood spots was set up with the quantitative covariates gas exposition time, slaughter weight and lean meat content as well as the covariates gas mixture, sex and day of slaughter. With this model, the h² was estimated to be 0.28 (P < 0.01). The best, although not genome-wide significant association signal for blood spots was identified on chromosome 7 at 8.8 Mb. A first inspection of the genome region revealed EDN1, encoding endothelin 1 (ET-1) as the most adjacent gene. ET-1 is a very strong endothelium-derived vasoconstrictor peptide and might be a promising candidate gene. These findings will now be validated using an enlarged sample and applying a fine-mapping approach.

Characterization of the relationship between olfactory perception and quantified compounds as phenotypes for genetic selection against boar taint using a recursive model

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Detecting and reducing boar taint is an important issue to maintain a high quality of fresh and processed pork. This deviant odor is mainly caused by skatole (SKA) and androstenone (AND) accumulated in fat and released when cooking pork meat. To avoid this, literature supports that breeding against boar taint is feasible considering the high heritability (h^2) of SKA and AND. However, carcass sorting at abattoir is performed by sensory evaluation yielding a human nose score (HNS). Sensory evaluation by trained assessors is considered a phenotype more closely related to consumer responses than chemical analyses. This study tried to characterize the origin of olfactory deviant attribution compared to SKA and AND concentrations by applying recursive mixed model (RM) using data of 1016 boar fat samples evaluated by 10 trained assessors. Some genetic correlations obtained by RM (-0.34 to 0.71 for SKA and -0.39 to 0.33 for AND) revealed that scores seemed to be assigned according to compounds related to SKA and AND in addition to these compounds themselves. Moreover, some heritabilities (h^2 0.04-0.20) from RM stayed moderate indicating that scores remained heritable without SKA and AND influence. Those results demonstrated the possibility to use HNS as boar taint trait but also the need to probably extend the “boar taint” phenotype to better relate it to the consumer’s perception for improving and applying HNS attribution at genetic level.

The effect of myostatin gene mutation on productive traits in Marchigiana young bulls in performance test
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Marchigiana cattle breed is an Italian local breed specialised for beef production. A double muscle phenotype occurs in this breed whose causative mutation (G>T) has been observed in MSTN gene, resulting in a GG normal genotype, a GT heterozygous one with an enhanced muscle development, and a TT hypertrophic genotype. Although the TT genotype is unwanted, the GT genotype is described in about 20% of population. It is questioned whether heterozygous animals show lower weight gains and performances, therefore the aim of this work was to confirm or confute this assumption by using performance records of 1074 young bulls tested at ANABIC genetic station (780 GG, 294 GT). The considered traits were morphological score, selection bull index, average daily gain index, muscularity index, average daily gain before and during performance test, weight at one year, muscularity. Data were analysed with ANOVA or Kruskal-Wallis tests. Results showed a significant increase in muscularity ($\mu\text{GG}=404.4\pm 62.3$, $\mu\text{GT}=444.6\pm 49.0$, $P<0.001$), muscularity index ($\mu\text{GG}=101.6\pm 9.7$, $\mu\text{GT}=105.1\pm 8.7$, $P<0.001$), and bull selection index ($\mu\text{GG}=102.2\pm 10.2$, $\mu\text{GT}=104.0\pm 8.9$, $P<0.01$) in heterozygous animals; no significant differences were observed for the other traits. Our results confirm the increase of muscularity related traits while other productive traits were not negatively affected. Funding: Italian Biodiversity Environment Efficiency Fitness— I-BEEF 2 — PSRN 2014-2020

Genetic parameters, genome-wide associations and single-gene effects for intramuscular fat content in a local dual-purpose cattle breed

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The aims of this study were to estimate genetic parameters, allele substitution and dominance effects of specific meat quality genes (CAPN1, CAST, DGAT1, FABP4, RORC, TG) and to perform GWAS for intramuscular fat content (IMF) in the endangered cattle breed Rotes Höhenvieh (RHV). In total, 542 RHV cattle were phenotyped for IMF and genotyped with the 777K SNP BovineHD BeadChip. Meat quality gene SNP were typed by DNA tests. Direct and maternal genetic parameters for IMF and weight traits were estimated using a multi-trait genomic animal model. The direct and maternal heritabilities for IMF were 0.76 and 0.29, respectively, while the direct-maternal genetic correlation was unfavorable (-0.18). Pronounced genetic variation and heritabilities for IMF indicated major gene effects, but only one SNP surpassed the Bonferroni significance threshold. Gene annotations based on the chromosome-wide significance level inferred candidate genes for direct (e.g. VWC2L, effect on feed efficiency) and for maternal genetic effects (e.g. GRHL2, effect on growth), but not the six well-known meat quality genes. Consequently, allele substitution effects for the variants of the six major genes were non-significant, probably due to the predominance of alleles. Related dominance effects were quite small, but the IMF values were highest in heterozygous animals for CAST and TG. Results indicate RHV peculiarities differing from commercial cattle breeds for effects of known meat quality genes, probably due to the specific breeding history. The gene annotations based on IMF genomic analyses suggest possibilities for meat quality improvement in the context of environmental adaptation.

Genotype by management-system interaction on origin group of milk fatty acids in Holsteins

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Cow health can be monitored using groups of milk fatty acids (FA) from de novo synthesis in the mammary gland. Environmental factors and management systems are known to contribute to the FA group. This study aimed to estimate genetic correlations among three management systems, i.e., tie-stall, free-stall, and pasture-based systems, for the FA group at early stages of lactation in Holsteins. A single-step genome-wide association study (ssGWAS) was also performed to detect regions unique to each management system. Data included over 1.45 million test-day records from 211,462 Holstein cows in their first or second lactation, calving in Hokkaido, Japan. Groups of “de novo” FA were predicted by Fourier-transform infrared spectroscopy of milk samples. The de novo FA percentage (DnF%) was calculated as the ratio of the predicted FA content to the total FA content for each origin group. The dataset was divided by lactation stage, defined as every 30 days in milk. A total of 119,822 animals were genotyped for 44,768 SNP. We used a tri-variate animal model with management systems as separate traits. Because of many genotyped animals, ssGWAS relied on the algorithm of proven and young (APY). The lowest genetic correlation was 0.96, between free stall and pasture, at stage 4 in the second lactation. At this lactation stage, we detected one significant region on chromosome 19 in all the management systems. The region was reported to be associated with milk FAs yield. Other lactation stages with genetic correlations greater than 0.99 among the systems were expected to have common significant SNP regions. More phenotypes and genotypes from multiple generations may be required to confirm the nonexistence of genotype by management-system interaction in the group of de novo FA at the early stage of lactation.

Investigating the effect of animal genotype on milk fat production in grazing dairy cows

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Milk fat composition and concentration are highly variable components of milk and while typically influenced by nutrition, an animal's genetic potential may also play an important role. The objective of this study was to investigate the effect of animal genotype on milk fat production in grazing dairy cows. Three groups of genetically divergent cows were evaluated within a large nutrition experiment where cows received a basal diet of grazed pasture and 3.6 kg of DM/cow/day of varying supplemental concentrates. The 3 genotypes evaluated were Purebred Jersey (JE; n = 60), Holstein-Friesian with high economic breeding index (EBI; Elite; n = 60) and Holstein-Friesian with the national average EBI (NA; n = 60). The study consisted of a 2-wk covariate and an 11-wk data collection period. Data were analysed using a repeated measures mixed model in SAS. The Elite and NA groups had significantly higher milk yield when compared to JE (26.5, 26.8, and 21.4 kg/cow/day, respectively; P<0.01). Milk fat concentration was highest for JE, intermediate for Elite and lowest for NA (5.74, 4.46 and 4.27 %, respectively; P<0.01). The Elite cows had higher protein concentration compared with NA (3.68 vs 3.53 %, respectively; P<0.01) but lower than JE (4.13%). Fat yield was lower for NA when compared to Elite and JE (1.14, 1.23 and 1.22 kg/cow/day, respectively; P<0.01). The Elites had significantly higher milk solid yields compared to NA and JE (2.19, 2.08 and 2.09 kg/cow/day, respectively; P<0.01). These results demonstrate the important role of animal genotype on milk fat production along with its ability to increase the economic and environmental sustainability of grazing systems.

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Colostrum yield is heritable and genetically correlated with immunoglobulins concentration in Holstein cows

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Bovine colostrum (BC) quality is given by immunoglobulins G (IgG) level, which should be at least 50 g/L for sufficient passive transfer of immunity in calves. 4 L of BC, ideally split into two meals, is the recommended volume in the first 12 h of life. However, calves often refuse to ingest 4 L of BC at birth. BC with high IgG concentration (> 50 g/L) enable delivering the same amount of antibodies (g) to calves with less volume, so that the high-quality BC in surplus can be stored for banking. Colostrum yield (CY, L) and IgG (g/L) are influenced by intrinsic and extrinsic factors in cows, but it is still unclear if these traits are genetically correlated and manipulable through genetics. Therefore, we estimated the heritability (h^2) of CY and IgG and their genetic covariance using REML approach. Data referred to Holstein cows' first milking, i.e. < 6 h from calving. For this purpose, more than 2,500 individual BC samples were stored to predict IgG via near-infrared spectroscopy with farmers recording for each sample the respective CY. IgG averaged 102 ± 33.62 g/L, while CY averaged 4.63 ± 2.28 L. The h^2 of IgG and CY was 0.22 and 0.07, respectively, and they were negatively correlated at both phenotypic (-0.26) and genetic level (-0.35). Results suggest that selection towards optimal quality and quantity of BC is feasible, but a proper index should consider their antagonistic association to ensure a genetic response in both traits in the right direction.

Heritability of traditional milk coagulation properties in Manchega dairy sheep

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This study aimed to estimate the heritability of traditional milk coagulation properties in the Manchega breed. Data from 791 Manchego ewes belonging to 5 flocks was used. The pedigree data was provided by AGRAMA. The rennet coagulation time (RCT), curd-firming time (k20), and curd firmness at 30 (A30) and 60 min (A60) were obtained using a lactodinamograph. Heritabilities were estimated using a univariate animal model that incorporated the parity type, age at calving-parity number and days of lactation as fixed effects and the flock-date as random effect. Marginal posterior distributions were estimated using Gibbs sampling with the GIBBSF90 program. Results were based on MCMC method consisting of 300,000 iterations with a burn-in period of 90,000, and only 1 of every 10 samples was saved for inferences. The heritability estimates (h^2) obtained were 0.16, 0.10, 0.14, and 0.10 for RCT, k20, A30, and A60 respectively. Overall, the h^2 of RCT and k20 were similar to those obtained in Sarda sheep, whereas the estimates of A30 and A60 were slightly higher. Our results suggest the possibility of using genetics for the improvement of the technological properties of milk within the breeding program. For that, it is necessary to study the genetic correlations of these traits with other traits already included in the breeding program in the Manchega breed. This research is part of Project PID2020-118031RR-C21/AEI/10.13039/501100011033.

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Effect of protein polymorphisms on milk composition, coagulation properties, and protein profile in dairy sheep

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The objective of this study was to quantify the effect of protein polymorphisms on milk composition, coagulation properties, and protein profile in dairy sheep from a New Zealand flock. Repeated test-day records from 147 lactating dairy ewes were obtained. Milk samples were analyzed for percentages of fat, protein, lactose, and casein; contents of α 1-, α 2-, β - and κ -casein, α -lactalbumin and β -lactoglobulin; milk coagulation properties, cheese yield, pH and heat stability. In the statistical analyses, two polymorphisms observed for α 1-, α 2-, β -casein, and β -lactoglobulin from HPLC chromatograms were included, along with three polymorphisms observed for α -lactalbumin. κ -casein was found to be monomorphic. Data were analyzed using a mixed model, including the fixed effects of ewe age, coat colour, litter size, stage of lactation, deviation from median lambing date, and the effects of protein polymorphisms, one at a time. Protein polymorphisms significantly affected milk composition and protein profile. Polymorphism of β -lactoglobulin significantly affected milk heat stability (AB>AA). Further studies on larger populations of dairy sheep are necessary to confirm the findings before applying genomic selection on polymorphisms.

Heritability of milk Fourier transform infrared spectra in sheep

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This work aimed at estimating the heritability of Fourier Transform Mid Infrared (FTIR) spectra of milk in sheep. Between 2020 and 2023, milk samples were collected fortnightly, at the a.m. and p.m. milking from December to July in an experimental flock of Sarda sheep. 1060 absorbance values from 925 to 5011 cm⁻¹ wavenumbers were captured with a MilkoScan FT+. After editing, 41075 spectra from 1256 ewes from 1st to 4th parity were retained. Variance components were estimated by a REML procedure (Asreml-R v4.2) applied to single trait repeatability animal models including the fixed effects of moment of milking, lactation stage within date and age-parity interaction; the random effects of the ewe within date, ewe within lactation and ewe across lactations. The individual genetic effect was included using the genomic relationship matrix (43390 SNPs). The genetic components of the covariance between repeated measures was nearly always higher than the other individual components. The average h² was 0.17 ± 0.10, ranging from 0 to 0.41 and being > 0.30 for 142 wavelengths. The highest values were in the intervals: 1141.968 -1273.14 cm⁻¹, related to amide III and phosphate bands and including the transitions in the vibrational energy of the triglyceride ester linkage C-O; 1442.892-1469.9 cm⁻¹, related to the vibrations of the CH₂ and -CH₃ groups; 2839.49 – 2978.38 cm⁻¹, associated with FA acyl C-H chain. Heritability estimates were low or close to 0 from 1624.22 to 1670.51 cm⁻¹ and from 3094.11 to 3653.52 cm⁻¹ where the absorbance due to the stretching of the O-H bonds (above~ 3000 cm⁻¹) or the H-O-H bending (~ 1600 to 1700 cm⁻¹), typical of water, occur. These results confirm that milk spectra can be used for the genetic improvement of innovative traits in dairy sheep.

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The genetic background of goat milk oligosaccharides content

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One of the most remarkable milk components are the oligosaccharides (OS). Milk oligosaccharides are complex carbohydrates which show a large diversity and are mainly undigestible. They can act as prebiotics and can selectively reduce the binding of pathogenic bacteria and viruses to the gut epithelium. Goat milk has higher concentrations of OS as compared to cow or sheep and has been suggested as a potential natural source of OS to produce infant formula. This study investigated the genetic background of goat milk oligosaccharide (gMOS) content by estimating heritabilities and performing a genome-wide association study (GWAS) based on 996 Dutch dairy goats. Goats were located on 18 farms in the Netherlands and were genotyped using the Illumina 70K goat chip array. The heritabilities for gMOS were high and ranged from 0.31 (3'-GL) to 0.85 (3-FL). The GWAS identified highly significant associations between five gMOS and SNPs located in 6 genomic regions. For 3'-NGL, 3'-SL and 3-FL cumulative QTL effects explained approximately 25% of the genetic variation and for 2'-FL this was more than 80%. Analysis of positional candidate genes showed that genes related to the metabolism of OS were significantly enriched, including genes involved in the transfer of L-fucose and sialic acid. Our findings suggest that genetic differences play an important role in determining the content of OS in goat milk and genotypes for a limited number of SNP can be used to identify goats with superior OS content.

Uncovering environmental effects and genetic parameters on the mineral composition of sheep milk

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The high levels of minerals in sheep milk affect its processing properties and impact product attributes and digestion kinetics. Their proportion in milk varies due to genetics, environmental conditions, and management. We assessed environmental factors influencing Ca, P, K, Mg, Na, Cl, S and estimated genetic parameters to identify their potential for selective breeding. Milk of 429 Comisana (CO) and 311 Massese (MA) reared at Asso.Na.Pa nucleus center was collected once during a total of 11 samples across a year. Milk was analysed for mineral, acidity and composition content and individual sheep information were also available. (Co)variance components were estimated using univariate animal linear mixed models. The additive genetic and residual terms were included as random effects while days in milk, parity and milk characteristics were considered as fixed effects. Heritability (h^2) estimates were moderate (0.24 to 0.33) for Ca, Cl, K, and Na in MA breed, and for K and Mg in CO breed. Higher h^2 was observed for Mg (0.59) and P (0.50) in MA and CO, respectively. Low h^2 was observed only for P in MA. The estimation of genetic parameters of milk minerals provides valuable information for breeding programs aimed at improving nutraceutical and technological properties at sheep population level.

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Poster 15

Molecular detection for *Coxiella burnetii* in bovine milk collected from bulk tank of small dairy farms in São Paulo, Brazil

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Coxiella burnetii is a gram-negative bacterium that causes a cosmopolitan zoonosis called Q fever. It infects a range of animals, but ruminants are considered the largest reservoir. It is known for causing gestational disorders in animals such as miscarriage, stillbirths and premature births, infertility, generating a large negative economic impact. Infection in humans occurs through inhalation of aerosols containing the bacteria, coming from the excrement, milk or birth products of these animals. The aim of this study was to research *C. burnetii* in 102 samples of milk from lactating cows, from bulk tank milk of small family farming from eight cities of Sao Paulo State, Brazil. The molecular techniques of Polymerase Chain Reaction (PCR), Nested PCR and real-time PCR (qPCR-Sybr using primers Trans3 and Trans4 and qPCR Probe using primers IS-202 and IS-249) were performed and then genetic sequencing was carried out. *C. burnetii* was detected in sixteen samples (15.68%) by Nested-PCR, twenty-two (21.57%) by qPCR/Sybr and thirty-two (31.37%) by qPCR/Probe. There was detection in seven of the eight cities tested (87.50%). There is evidence of possible outbreaks and negative impacts on livestock that the presence of *C. burnetii* can cause silently, as notification and testing of the bacteria is not carried out.

Single-step genomic prediction for live animal ultrasound measures of carcass traits in Czech Aberdeen Angus

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Although meat quality is still not reflected in the price of meat at commercial abattoirs in the Czech Republic, many Aberdeen Angus breeders prefer animals with better meat quality. These are mainly producers who implement a system of selling meat from farms or running their restaurants. In this case, customers are more likely to demand high-quality meat. For this reason, in 2019, regular annual live animal ultrasound measurements of meat quality-related traits were launched. Four measurements are collected – rump fat thickness (P8FT), rib fat thickness (RBFT), eye muscle area (EMA), and intramuscular fat content (IMF). In addition, the weight at scanning (SCW) is recorded. This study aimed to develop a genomic evaluation for live animal ultrasound measures and investigate the impact of using genomic information on genetic evaluation. Ultrasound measures were collected from 2,500 live animals, the relationship matrix included 7,251 animals, and 2,278 animals were genotyped using GeneSeek GGP Bovine 150K, BovineSNP50 v3 BeadChip, and EuroG MD beadchips. Common SNPs located on all chips were used for genomic evaluation. The number of effective SNPs was 32,126. BLUPF90+ software was used for analysis. Firstly, the genetic parameters were estimated using AIREML. The obtained heritabilities were 0.37 ± 0.05 (SCW), 0.38 ± 0.05 (P8FT), 0.34 ± 0.04 (RBFT), 0.27 ± 0.05 (EMA), and 0.12 ± 0.04 (IMF). The genomic breeding values (GEBVs) were predicted using the ssGBLUP method. Correlations between BVs (predicted earlier) and GEBVs were 0.9–0.98 for non-genotyped animals, 0.9–0.95 for genotyped animals with records, and 0.82–0.85 for genotyped pedigree animals. No significant changes in reliabilities for non-genotyped animals were found. However, GEBV's reliabilities for genotyped animals were distinctly higher than BV's. For genotyped animals with records, the reliabilities on average increased by 0.29, 0.21, 0.2, 0.24, and 0.16. The increase was even higher for genotyped pedigree animals by 0.44, 0.4, 0.39, 0.36, and 0.27 for SCW, P8FT, RBFT, EMA, and IMF, respectively. These results confirm the benefits of single-step genomic evaluation. This study was supported by projects QL24010025 and MZE-RO0723.

Session 44

Poster 17

Evaluation of texture properties and consumer perception of pork from high-marbled Duroc leptin receptor genotypes

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Intramuscular fat content (IMF) and marbling play an important role in pork eating quality, although their impact on overall acceptability largely depends on the consumer habits and market segment. Even among lean eaters, acceptability increases with IMF, but not with visible fat. Expected and experienced quality are two different quality cues that can be antagonistic, since, while IMF contributes to enhance experienced hedonistic quality, an excessive perception of fat may undermine consumer expectations. Using data from a Duroc line devoted to produce premium pork products, we have previously shown that the homozygous recessive for the T allele of the missense polymorphism rs709596309 C>T in the leptin receptor gene (LEPR) increases IMF and marbling. Here, we present the results of an experiment conducted to investigate whether (1) a high level of IMF and (2) the LEPR genotype contribute to pork quality differentiation and affect consumer preferences. Instrumental quality attributes and consumer preferences were evaluated on 48 purebred Duroc (24 LEPR-TT and 24 LEPR-C) and 24 Duroc-25% crossbred loins by means of a texture profile analysis and a household Home Use Test. High-marbled purebred Duroc (7% IMF) showed better texture and sensory acceptance than Duroc-25% (2.1% IMF). For purebred Duroc, no influential differences were observed between IMF levels (9.5% vs 5.5% IMF), nor between the two LEPR genotypes. It is concluded that consumers clearly differentiate between purebred and Duroc-25% crossbred, but not between levels of IMF higher than 5.5%. However, a potential effect of non-purchasing consumers in favour of the LEPR-C genotype was detected.

Estimation of individual CH₄ emissions using fecal near infrared spectra for young, dairy and beef cattle
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Last years several indirect methods/proxies have been investigated to predict individual enteric CH₄ emissions from cattle without using reference measurement methods which are expensive and time-consuming. However, there is a lack of proxy allowing to take into account individual variabilities for non-lactating animals: young cattle, dry cows or beef cattle. Faeces analysed by near infrared (NIR) spectrometry are relevant to consider because such spectral information has already demonstrated their interest to predict forages dry matter intake and digestibility. Otherwise, CH₄ emissions can also be correlated to these last parameters. A dataset of 851 reference values has been constituted including CH₄ values measured with Greenfeed system averaged on 14 days (274±135 g CH₄/day) and corresponding spot fecal NIR spectra (sampled on day 14). 256 data were from lactating cows (Holstein), 249 data from young cattle (Holstein heifers, young Belgian blue bulls) and 346 data from beef cattle (Charolais, dual purpose Belgian blue, Belgian blue cows, beef-on-dairy crossbred bulls). These categories of animals have been used to develop independent and common prediction models (10% of animals per category discarded for independent validation (P)). The PLS model including all categories of animals showed the best statistics: RPD=3, R²P=0.87 and SEP=48 g/d, demonstrating the great potential of this approach even if the robustness needs to be improved.

Session 45

Theatre 2

Learning behavior of dairy cows in automatic milking systems

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The number of farms using automatic milking systems (AMS) is increasing continuously. Hence, improving cow behavioral response to technical environments (cow-AMS interactions) becomes increasingly important for smooth cow traffic and welfare. This response to technical environments is largely determined by learning behavior. As AMS are crucial for daily routine of dairy cows, they are suitable test systems for behavioral studies. Aim of the study was to quantify cows' learning behavior, using traits defined as average daily AMS visits, milkings and rejected milkings. A key factor in this context is the time period a cow was familiar with the AMS. AMS data from 13 German dairy farms were analyzed. Data comprised 7,803 Holstein cows with 19,259 lactations documented from February 2009 to August 2023, and was aggregated on a per-day basis to form 5,074,564 daily cow-AMS interactions. Stage of lactation was defined as three time periods for days in milk (DIM) 0-99, 100-199 and 200+. Time periods for learning (familiarity with AMS) were defined as 1-25, 26-50, 51-75, 76-100, 101-150, 151+ days. Analyses were conducted in SAS using a mixed linear model with fixed effects daily milk yield and combined effects of lactation (1, 2, 3, 4, 5+)*familiarity with the AMS, and farm*season. Cow effects were included as random. Both daily AMS visits and milking frequencies significantly declined with age and familiarity with AMS. Learning takes place mainly during the first 25 days at AMS and is strongest in heifers during DIM 100-199. Genetic analysis including genomic data is ongoing, and will reveal a better understanding of genetic mechanisms influencing learning.

Identification of SNPs affecting conformation traits of dairy cows

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This study aimed to identify single nucleotide polymorphisms (SNPs) present in microarrays associated with conformation traits of Polish Holstein Friesian (PHF) cows. Genomic data were obtained during routine estimation of breeding value with the use of customized SNP arrays, data on cow conformation traits was recorded by Polish Federation of Cattle Breeders and Dairy Farmers. A total of 376 PHF cows were examined. Following data cleaning, 41,705 SNP records were considered for statistical analysis. Additionally, 56 variables related to cow conformation traits were analysed. Statistical analyses were conducted using the Bigstep analysis method implemented in the R software package. The results showed that certain SNPs significantly ($P < 0.001$) influenced specific conformation traits. 4 SNPs from chromosomes 2, 6, 24 and 29 (UMD_3.1 reference genome) affected Udder position; 3 SNPs Hoof angle (Chr9, 16 and X) and Locomotion (Chr 2, X); 2 SNPs impacted the following traits: Depth of the body (Chr5, 14), Rear leg stance (rear view) (Chr3, 15), Rear teat placement (Chr1, 19), Suspension of the fore udder (Chr1, X) and Width of the udder (Chr7, 13). 1 SNP had a significant impact on Front teat placement (Chr7), Height at the withers (Chr24), Hindquarters' conformation (chr16), Legs and hooves (Chr16), Rear leg stance (side view) (Chr19), Udder (Chr8). The analysis underscores the utility of microarray data for investigating associations between genes included in the arrays and conformation of cows' traits, which may hold promise when combined with performance analysis for exploring optimal conformation traits in cows predisposed to specific milking types. Financially supported by the Minister of Science under the program "Regional Initiative of Excellence" (RID/SP/0017/2024/01)

Optimize the individual insemination strategy in extended lactations of dairy cattle using a neural net to predict phenotypic lactation curves

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Expanding the voluntary waiting period until the start of insemination in dairy cows is a frequently discussed topic. Different benefits in fertility, health or management and therefore economics are expected. The management of extended lactations raises new questions, e.g., when to start insemination. We propose an approach and tool to help answering these questions. Our approach can be divided into two tasks: first, predict an individual phenotypic lactation curve; second, use the predicted curve to calculate the best time for insemination, depending on the mean of first service to conception interval and the farm strategy for dry-off level and duration. To predict a phenotypic lactation curve at the beginning of a lactation, we use general information of the animal, e.g., breed, lactation number as well as milk yield information. We also use information at the farm level, e.g., the mean herd-test-day effect in last year, which is a fixed effect from the routine breeding evaluation in milk traits. With all this information of approximately 3.1 million lactations of German dairy cows born from 2017 onwards, we train and validate a feedforward neural network by minimizing the mean squared error of the prediction and the real shown lactation curves based on test day measurements. The results from a validation scenario show that our model's predictions deviate by approximately 4 kg milk per test day, which from our perspective, is considered a good value given the variations observed in the test day measurements within the lactation. Finally, we have integrated the individual animal insemination recommendation into a tool to make it usable for dairy farmers in daily work.

Real-Time Milk Yield Prediction and Health Monitoring System for Dairy Cows

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Ensuring the sustainability of the dairy sector largely depends on effective health and welfare monitoring of the cows. Milk yield is a crucial indicator for productivity, with deviations often signaling underlying issues such as disease or metabolic status changes. While numerous milk yield deviation models exist, their on-farm real-time validation remains limited. We present a novel approach that adapts an established linear mixed model for real-time prediction of quarter-level milk yield. Our method integrates statistical process control to identify deviations. To this end, we developed a fully automated data pipeline that collects data from dairy farms, reports insights to farmers through a dynamic web application, and facilitates seamless feedback collection on clinical symptoms and treatments. We analyzed the detected milk yield deviations for distinct categories: deviations with clinical signs reported by farmers, deviations with subclinical signs as per deviating values in different sensor data (e.g., electrical conductivity), changes in management (e.g., feed alterations), natural fluctuations, and potentially undetected disease cases validated against treatment records. In a 3-month trial phase, from 177 detected deviations, 23 were cases with clinical signs, 44 with subclinical signs, 31 were due to a change in management, and 79 were due to issues with the milking process or natural fluctuation. By leveraging real-time predictive modeling and comprehensive health monitoring, our approach allows dairy farmers to proactively manage herd health, mitigate productivity losses, and ensure increasingly sustainable animal welfare.

Session 45

Theatre 6

Modern Genetic Evaluation Systems: A Python-Based Programming Approach

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The field of animal breeding is continually advancing with the development of new technologies and methodologies for data collection and analysis. As the industry progresses, modern programming approaches can ensure genetic evaluation systems (GES) stay up to date. Python is a fast-developing programming language with scientific computing libraries (NumPy, SciPy). This study evaluated Python's (v3.12.1) practicality in developing a GES. AlphaSimR was used to simulate a dataset mimicking the Angus breed, including pedigree, phenotypes (birth weight (BWT), weaning gain (WG), post-weaning gain (PWG)), and true breeding values (TBVs) for 976,400 animals over 15 generations. Estimated breeding values (EBVs) were predicted using pedigree-based BLUP and a multiple-trait animal model. The preconditioned conjugate gradient iteration on data algorithm was used to solve the mixed model equations. Convergence was defined as the residual vector norm divided by the right-hand-side norm and the convergence threshold was 1×10^{-8} . The software performance was assessed on a Linux server with an Intel(R) Xeon(R) Gold 6242 CPU @ 2.80GHz, 96 GB of RAM, and eight cores. Accuracy was calculated as Spearman rank correlation between TBVs and EBVs. Estimation of breeding values required the most computing time, 79 minutes. However, multiprocessing reduced the run time to 29 minutes. Prediction accuracy was 0.86, 0.78, and 0.70 for BWT, WG, and PWG, respectively. Python seems an ideal language for a modern GES, providing results within a reasonable time frame, however updates to optimize performance should be applied for further efficiency.

Immunomodulatory effect of small extracellular vesicles from the plasma of heat-stressed cows on bovine immune cells

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Warm weather has been an environmental problem faced worldwide lately. From the animal's welfare point of view, hyperthermia affects important functions of immune cells. Small extracellular vesicles (sEVs) are responsible for cell-to-cell communication. They may also regulate immune responses by realizing their cargo in the target cell, which is affected by outer stress. The objective of this research was to examine the immunomodulatory impacts of plasma sEVs isolated from cows experiencing two distinct conditions: thermal comfort (TC, THI <68) and a natural four-day heatwave (hyperthermia, HS, THI_{max}=84, THI_{min}=69), on bovine monocytes (CD14+) and PMNs. The sEVs were isolated from the blood of three healthy Holstein cows under both TC and HS conditions. During TC conditions, Monocytes (CD14+) and PMNs were purified from the blood of 6 clinically healthy Holstein cows. Monocytes were sorted using Magnetic Activated Cell Sorting and PMNs using a Percoll gradient isolation. Cells were then co-cultured with sEVs. Statistical analysis was conducted using SAS® (SAS Institute Inc., Cary, NC, USA). General results showed that the chemotaxis (P= 0.0204) and phagocytosis (P = 0.015) capabilities were significantly affected when leukocytes were cocultured with HS sEVs, compared to TC sEVs. In summary, sEVs released during heatwave conditions impaired the ability of immune cells to phagocytize, which could elevate cows' susceptibility to infections.

Session 45

Theatre 8

Coupling remote sensing techniques and Virtual Fencing in Alpine dairy production system: enhancing livestock management and environmental conservation

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Pastoral systems are a valuable source of income for mountain farmers, while providing multiple ecosystem services. Proper management of these systems is crucial for their conservation and for the sustainable use of natural resources. Virtual Fencing (VF) is a promising technology that can help to achieve these goals. It consists of a GPS collar that can confine animals to a grazing area defined from remote, by delivering a coupled aversive stimulus (i.e., an acoustic warning followed by an electric pulse). The objectives of the study were (i) to evaluate the animal response to VF and (ii) to test the feasibility of coupling remote sensing (RS) imagery with VF data to estimate animal feed intake in a mountain environment of dairy system (alpage). Our study was conducted in the Eastern Italian Alps (Mount Zoncolan, IT, 1600 m of altitude, with different pastoral typologies), where VF was installed on 35 Bruna dairy cows managed both summer pastures and on ski slopes, with daily increases in the virtual grazing areas. The results showed that the animals responded well to both types of pasture areas. Furthermore, by analyzing the spectral response of the vegetation (i.e. calculating the vegetation index NDVI or the LAI parameter) between pre- and post-grazing, information on their feed intake was estimated. This study showed the potential of coupling VF and RS data in the alpage environment, where in-situ monitoring is often inaccessible and costly, contributing to tackle the abandonment of this area during the summer.

The confidence ratio: an approach to calculate the confidential learning of the virtual fencing system
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Virtual fencing (VF) is a tool to effectively manage grazing cattle. The visual cue of a common fence is replaced with an audio cue (AC; a melody rising in pitch) emitted by a GPS based VF-collar when the animal approaches the virtual boundary. When the animal continues moving forward, a short-time electric pulse (EP) follows the AC after the last note of the melody. Learning the association of the AC and EP is a prerequisite for an adequate interaction with the VF. Several studies used a success ratio to assess learning, to calculate the proportion of AC with no subsequent EP out of the total number of AC. However, the frequency of the animals' voluntary interactions with the VF is not considered in this approach with a consequent lack of understanding. A high success ratio reveals either a high number or a low number of AC. For example, the success ratio for an animal receiving one or 14 AC without the subsequent EP is both the maximum '1'. Therefore, we multiplied the result of the success ratio with the number of AC divided with 20 to weight these different cases and to express the confidential interaction with the VF in a ratio between '0' and '1'. We choose 20 as the maximum number of AS based on the integrated mode change function of the Nofence VF collars which changes from teach to work mode when the animal received the 20th acoustic signal without a subsequent electric pulse. Data from a 12-d training period with 16 heifers were used to test this approach. It appeared to reflect a confidential learning of the VF system.

Session 45

Theatre 10

Effectiveness of energy dispersive X-ray fluorescence (ED-XRF) for the prediction of mineral elements in individual cow urine

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Mineral elements, along with proteins, free amino acids, and sugars, represent one of the main indicators of heat stress in dairy cows urine. For these reasons, there is interest in the development of novel techniques aimed to predict urine mineral elements in reasonable time and low costs. The objective of the present research was to assess the effectiveness of energy dispersive X-ray fluorescence (ED-XRF) for the prediction of Ca, K, Mg, Na, P, S and Cl in dairy cows urine. Individual urine samples were collected from 45 Holstein Friesian cows in 9 commercial dairy farms located in north of Italy. One aliquot was used for mineral extraction followed by ICP-OES analysis. The second aliquot was used for ED-XRF analysis on untreated urine samples. The training of the ED-XRF method was performed on a subset of 30 individual urine samples, as a regression equation between mineral elements obtained through ICP-OES analysis and normalized impulses profiled by ED-XRF instrument. Instead, the testing of the ED-XRF method was performed excluding samples used in the training dataset and considering only the remaining samples (n = 15) as a regression equation between minerals measured through ICP-OES versus minerals predicted through ED-XRF using the training model. Results highlighted moderate accuracy of the ED-XRF technique for the quantification of Ca, with coefficient of determination in testing (R²) of 0.60 while greater accuracies were obtained for the quantification of the other elements, with R² ranging from 0.86 (Mg and K) to 0.99 (Cl).

Effects of unplanned regrouping on activity, feed intake and milk yield of dairy cows

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Regrouping is a common practice in commercial dairy farms, still it causes stress for the animals affecting their behaviour. This analysis focuses on events of unplanned regrouping caused by a malfunctioning selection gate on a research farm in Germany. Because of incorrect selection after milking some animals end up in an unfamiliar group for 12h until next milking. In three equally designed groups with 64 animals each, 82 events of unplanned regrouping of 71 animals were observed in 6 weeks (October 2022, February 2023). Activity (ACT), dry matter intake (DMI) and milk yield (MY) were compared on the day before and after regrouping (RG 0/1) using linear mixed models in SAS 9.4®. Fixed effects were week, RG, lactation number (LN: 1: 1, 2: 2, 3: ≥ 3), lactation day (LD: 1: <105, 2: 105-210, 3: >210), body weight and dominance index (BW and DI: 1: <mean-standard deviation(sd), 2: mean-sd – mean+sd, 3: >mean+sd). The animal was included as random effect. ACT was significantly ($p < 0.05$) affected by RG (0: 30.07±2.12 (LSMean±standard error), 1: 39.02±3.48). DMI differed significantly ($p < 0.05$) for animals in LD 1 compared to LD 2 and LD 3 (1: 20.41±0.62, 2: 23.01±0.67, 23.80±0.71) and for RG (0: 22.91±0.56, 1: 21.90±0.56). MY differed significantly ($p < 0.05$) for animals in LN 1 compared to LN 3 (1: 34.04±1.12, 2: 37.42±1.92, 3: 40.21±1.44) and for DI (1: 33.41±1.38, 2: 37.18±1.08, 3: 41.08±1.54). For RG, MY showed a trend (0: 37.61±1.02, 1: 36.83±1.01, $p = 0.066$). The results show, even short periods of regrouping can affect behaviour and MY. This analysis will be carried on to extend the observation days and to compare the results to planned regrouping.

Session 45

Theatre 12

Insights into monitoring livestock on the move

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Beef production involves the transportation of cattle to various locations for sale, finishing, and slaughter. These journeys expose animals to a range of stressors associated with curfews, novel environments, and handling, including fear, fatigue, dehydration, and metabolic compromise. Long-haul road transport is common practice in Western Australia with northern rangeland beef cattle subjected to journeys over 2,400km to reach processing facilities in the south, yet studies on the impact of such duration of travel is lacking. Australian federal regulations for adult cattle stipulate a maximum time off water to be 48 hours. This study developed a protocol for monitoring cattle undergoing transport and aimed to evaluate the effect of rest durations on welfare, using assessments of behaviour, health, demeanour, physiology, and meat quality. *Bos indicus* cattle ($n = 952$) were assigned to short, medium, or long rest periods after approximately 36 h transport in a commercial setting. Using Factorial ANOVA, preliminary results of behavioural analysis at the rest stop and in lairage indicate cattle given long (~64 h) rest periods had started to express normal patterns of maintenance behaviour (eating, drinking ruminating) compared to those given a short (~18 h) stay. Post-slaughter measures of blood markers analysed by GLMM indicated that most cattle had a physiological stress response, e.g., higher neutrophil count, which was more pronounced in the short treatment group ($p < 0.05$). Additionally, urine was less dilute for cattle in short compared to long rest groups ($p < 0.001$). The duration of stay had minimal impact on meat quality. Results suggest a rest period of ~64 h off-truck may permit recovery from transport stress and optimise cattle welfare after long haul journeys.

Development of a multidimensional and customizable decision support tool for dairy culling management
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The economic performance of dairy farms essentially relies on the farmer's ability to select the most suited cows for their production system. A major constraint lies in the time required for data encoding and analysis at the farm level. Our solution offers a data centralization service to facilitate its access and use. The study seeks to develop an innovative decision support tool (DST), named Win/Lose, which ranks dairy cows according to their economic profitability by integrating various data such as milk production, veterinary costs and metabolic disorders. The DST aims at identifying cows to keep (Win) and those to cull (Lose), thereby enriching the farmer's decision-making capacity. The DST calculates a profit index based on milk yield, fat content, protein, culling weight, somatic cell count, and their respective net unit margins. The index also incorporates fertility performance. Through predictive models utilizing the mid-infrared spectrum of milk, the DST analyzes data on fatty acids, cheese-making properties, minerals, energy balance, infections, and methane production. This multi-dimensional approach refines cow selection, offering customization according to the farmer's priorities. This tool enables objective selection, providing farmers with a broad range of indicators for informed decision-making on cow culling and complementing the economic and environmental efficiency of their operations. The use of the DST contributes to a significant improvement in the profitability and sustainability of dairy farms. By streamlining data analysis and customizing selection criteria, this tool gives farmers the freedom to define their production strategy, aligned with their economic and ecological objectives.

Session 45

Poster 14

Genetic correlations between udder conformation and milking traits of Holstein-Friesian cows milked by milking robots

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Our study aimed to recognize the genetic background of those relationships based on genetic correlations (rG) between the selected udder conformation traits (UCT) and milk yield (MY), milking characteristics (milking efficiency (ME)), teat cup attachment time (AT), and number of attempts (AN). The study covered 796 Polish Holstein-Friesian cows milked with automatic milking system (AMS). The primiparous cows were assessed for: fore udder attachment (FUA), front teat placement (FTP), teat length (TL), udder depth (UD), rear udder height (RUH), central ligament (CA), rear teat placement (RTP), and rear udder width (RUW). In order to estimate the genetic correlations between the udder dimension traits and milking traits, the GIBBS1F90 program and a linear two-trait animal model were used. The number of samples in the burn-in period was determined using the POST-GIBBS1F90 program. The results of our study revealed that the rGs between individual UCT and MY are weak to moderate (-0.003 to -0.36). The rGs between the UCT and milking characteristics were weak to strong (in the range of -0.25 to 0.20 for ME, -0.60 to 0.84 for AN, and -0.57 to 0.98 for AT). In both cases, there are positive and negative rGs. The positive rGs for AN and AT were negative for ME simultaneously (TL, RUH, RUW, and CL). When the rGs were negative for AN and AT, they were positive for ME (UD, FTP, and FUA). The detailed knowledge of genetic relationships between individual UCT and ME and milking characteristics in AMS seems to be a valuable tool for making better breeding decisions on farms equipped with AMS. Financially supported by the Minister of Science under the program "Regional Initiative of Excellence" (RID/SP/0017/2024/01)

The effects of bed cleanliness on the lying postures of Japanese Black fattening cattle

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Background Lying behavior plays an important role in comfort of cattle around resting. Cattle frequently changes their head or leg positions while lying. As bed cleanliness is associated resting comfort of cattle, investigating the effects of bed cleanliness on these postures might provide more clues on resting comfort. The study aimed to investigate the effects of bed cleanliness on lying postures of fattening cattle. **Methods** The study was conducted at deep-bedded cattle shed in March 2023. We observed lying postures of 8 Japanese Black fattening cattle for 24h before and after bed cleaning. We systematically classified 27 possible lying postures based on head and leg positions. Head positions were classified into L (lifted), T (turned backward), and E (extended). Number of extend front and hind legs (0,1,2) were also counted. Duration and frequency of each posture in cleaned and uncleaned condition was compared with t-tests, and the proportion of each posture transitions were compared using fisher's exact tests. **Results** Frequency of 14 postures were expressed statistically significant ($p < 0.05$). Duration of lying postures with head lifted, both legs tucked (L00) increased in cleaned compared with uncleaned condition ($p < 0.05$). Hence cattle show longer L00 on clean bed surface. Moreover, there were increase in frequency ($p < 0.05$) and duration ($p < 0.05$) of postures with head turned backward, both legs tucked (T00). Proportion of transition from L00 to T00 and from L00 to T01 increased in cleaned condition ($p < 0.05$). These results indicate a cleaner resting place also promotes the expression and transitions of T. Our study suggests that lying postures with legs tucked and postures with heads turned backward are relevant with resting comfort of fattening cattle.

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Poster 16

Studies on in vitro Embryo Splitting as a method for Sex-Selective Production of cattle

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This study was conducted to increase female calf production through the use of embryo splitting technology and BTMs (blastomeres) for genetic testing, specifically for sex determination. In vitro fertilized (IVF) embryos are produced from ovaries of slaughtered cows. After 26 and 50 hours of IVF, 2-8 cell stage embryos are selected, treated with 0.1% pronase to remove the zona pellucida, and split by pipetting. BTMs are cultured for 7 to 8 days (IVF=day 0). Injected hCG on the day before transplantation to synchronized recipients, and transplanted blastocysts cultured from 2-cell, 5-cell, and 6-cell embryos. Pregnancy was confirmed on day 40, using a pregnancy test kit (IDEXX, USA). The cleavage rate of BTMs (blastomeres) was significantly higher in split BTMs (SB) from the 2-cell (83.3%) and 4C-SB (69.1%) split at 26 hours after IVF ($p < .01$). The blastocyst developmental rate was notably higher in 2C-SB (36.1%) split after 26 hours ($p < .01$). After fixation in 3.7% paraformaldehyde, the total cell number was examined by nuclear staining with DAPI. Blastocysts derived from BTMs were significantly fewer than untreated blastocysts (control). This study affirms that the most efficient stage for sex-specific embryo production is the 2-cell stage 26 hours after IVF. When blastocysts derived from separated embryos at the 2-cell, 5-cell, and 6-cell stages were individually transplanted, successful pregnancy was observed with the blastocyst from the 5-cell stage. In this case, the blastocyst from the 5-cell stage exhibited a distinct inner cell mass (ICM) and a high total cell number. Therefore, the total cell number of the blastocyst may influence the success of embryo transplantation.

Effect of Milking pulsators efficiency on Udder Health

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Among the machine-related factors influencing milking performance and udder health, proper functioning of the milking pulsator (PT) is crucial. Correct pulsation prevents congestion and edema in the teat tissue during milking. This study aims to investigate the effect of malfunctioning PTs on udder health by evaluating milk SCC during the lactation (DIM). In 2023, about 25,000 PT tests were conducted using VaDia® on 1,255 farms and about 50,000 cows. The recorded PT parameters included: rate, ratio, limping, b-phase, and d-phase. Out-of-range PT parameters were identified by threshold criteria according to ISO 5707:2007. Three farm classes were defined based on the percentage of PTs device with, at least one, parameter out of range: < 20% (Class A), between 21% and 60% (Class B), > 61% (Class B). The individual SCC linear score (SCC-LS) was calculated from official DHI. Data were analysed by a mixed linear model. The interaction SCC-LS and farm classes resulted significant (P-value < 0.01). From 30 to 150 DIM, the percentage of cows with SCC exceeding 200,000 increased from 1.36% (Class A), to 6.56% (Class C). Our results indicate that malfunctioning pulsators cause the increase of SCC in the first lactation phase deteriorating the milking performance and raising the risk of machine-related udder harms.

Session 45

Poster 18

Genetic identification to ensure blockchain-based traceability in beef supply chain

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The objective was to assess blockchain-based traceability in the beef supply chain using genetic markers. A total of 250 beef cattle from two farms located in Veria, Greece, were randomly selected. Animals were slaughtered in designated local facilities. Farm, animal, and meat traceability data were collected and a decoding system using QR-coding was developed. Individual blood samples were collected from the jugular vein in EDTA vacutainers during slaughtering process. From each carcass, a meat sample was obtained at the meat processing unit in sterile tubes using a disinfected knife. DNA was extracted from each blood and meat sample using designated kits according to manufacturer recommendations. DNA samples were genotyped for 17 microsatellite markers, recommended by the International Society for Animal Genetics; BM1818, BM1824, BM2113, CSRM60, ETH10, ETH185, ETH225, ETH3, ILST006, INRA005, INRA023, INRA063, SPS115, TGLA122, TGLA126, TGLA227, TGLA53. Genotyping has been completed for 142 pairs of samples (blood and meat) that were found to be correctly assigned. Preliminary genetic identification results suggest that blockchain technology could ensure traceability in the beef supply chain from farm to fork. This research was co-financed by the European Regional Development Fund and Greek National Funds through the Operational Program of Central Macedonia 2014-2020 (KMP6-0219662; AuthenBeef).

Automated measurement of milk yield in suckler cows

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The productivity of suckler cow farms depends on the number of calves weaned per cow and of their weights at weaning, as these are the criteria used for selling them. To ensure calf weight gain, supplementation with concentrates has become the preferred method, therefore, it is essential to maintain and develop the dairy potential of suckler cows (SC) to reduce feed costs. The aim of this project is to develop a validated reference model for predicting milk production (MP) in SC. The standard method for quantifying MP involves weighing the calf before and after suckling. This method is time-consuming and requires significant manpower, and is therefore only used in a few experimental stations. The development of automated weighing and electronic animal identification has led to the emergence of an alternative method that could become the reference. The concept is to allow the calf to suckle its mother freely, with a compulsory weighting between its pen and mother one. The software processes the weights collected automatically to estimate MP. INRAE's Herbipôle unit designed and tested a prototype 7 winters on 117 cows, 71 Charolais (CH) and 46 Salers (SA). Each winter was divided in 6 periods of 14 days. Each period alternated between 4 days of standard method (2 sucklings/day) and 10 days of free suckling with the device based on automatic weighing at each pen change. An algorithm transform calf weights into MP. The calves suckled 3.1 ± 1.5 times/day during the first period compared to 4.5 ± 1.7 during the last. The free device estimated that MP was 8.3 ± 2.1 kg/d for SA, 6.9 ± 1.3 for CH, with a good correlation with the reference method (r^2 from 0.86 to 0.66 from period 1 to 6). The free device has been validated and will be extended to various suckler cattle experimental stations to measure MP of other breeds in various breeding conditions.

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Poster 20

Behaviour and welfare monitoring of cattle using computer vision and machine learning

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A profound knowledge of cattle behaviour allows to jointly optimise animal welfare and productivity. Currently, behaviour monitoring of cattle is mostly performed using accelerometers, either or not in combination with localisation based on GPS or ultra-wideband technology. Essential cues for animal behaviour, such as social interactions, largely remain hidden for these sensors. To this end, we developed a computer vision pipeline consisting of four building blocks: pose estimation, animal segmentation, animal re-identification and behaviour prediction. Various state of the art computer vision technologies were combined to cope with specific challenges at commercial farms. As such, our pose estimation algorithm uses an adaptive Kalman filter based keypoint tracking algorithm to improve the consistency of keypoint detection and lowering the computational cost of the pipeline. We were able to detect up to 80% of the ground truth keypoints with high accuracy, with only a limited drop in performance for nightvision recordings. Moreover, the temporal consistency of generated keypoint coordinates and derived skeletons was largely improved. Our re-identification algorithm, on the other hand, has built-in versatility towards the pose and orientation of the animals. This offers the possibility to re-identify cattle anywhere and anytime, in contrast to state of the art algorithms. The obtained rank-1 accuracy of the re-identification is 81.6%. With this pipeline, we can analyse the animals' behaviour synchronicity, study the location preference of the animals and investigate the social interactions and network in cattle.

Climate change indicators as proxy for economic efficiency of dairy farming

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Decoupling of operational indicators led to optimization of economic performance and desirable outputs while minimizing environmental impacts, undesirable output and ecosystem disservices of farms. Environmental indicators can be easily obtained with simplified life cycle assessment whereas economic indicators are more difficult to obtain both for privacy reasons and for lack of records at farm level. Satisfactory economic and environmental performances are both associated to efficient use of resources. The aim of this study was to assess the correlation among economic and environmental indicators in dairy farms. A number of 52 dairy farms located in Arborea (Or, Sardinia Italy) was surveyed with face-to-face interview following a Life Cycle Assessment procedure for determination of Climate Change impact (CC in kg of CO₂ Eq/kg of FPCM) and economic records were also gathered. Life Cycle Assessment procedure was certified for CC, according to ISO 14040 and 14044. Milk prices was 0.56 €/liter. Income over feed cost and added value (revenues from ilk and meat minus feed costs, services and fuel) were calculated. The data collected were representative of the last 3 years (2020-2023). Farms had 178 milking cows (range 46 to 470), delivering 9709 kg/yr (range 6301 to 1228) of Fat and Protein Corrected Milk (CPCM) per cow, 74 Ha (range 26 to 216) and represented more than 50% of the milk delivered to the Arborea Cooperative (3A). All farms were intensively managed with double crop plan ryegrass-corn silage and used to buy off-farm most part of concentrates. CC, 100% allocated to milk, was equal to 1,38 kg of CO₂Eq/kg of FPCM (range 1.8 -1.1) with a breakdown of 41.4% in enteric methane, 15.6% from manure, 5% from produced feeds, 32.7% from purchased feeds, and 5.5% from energy and fuel. IOFC was equal to 3516,00±952,58 €/yr per cow. CC and IOFC were negatively correlated ($r=-0.62$). The maximum levels of added value can be reached only below certain levels of CC. An output (€ of added value/yr per cow) Vs output (CO₂eq/kg of FPCM) production frontier was calculated as indicated by the line €/yr per cow of added value = $-4189 \cdot CC^2 + 10418 \cdot CC - 4534$.

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Poster 22

Effects of an automatic feeding system on Swiss dairy farms

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Agricultural businesses in Switzerland are growing steadily. As a result, the workload is also increasing. At the same time, the performance of dairy cows is increasing, which makes feeding the animals according to their needs a major challenge. The aim of this study was to analyze the effects of an automatic feeding system on Swiss dairy farms. A total of 5,919 daily milk yields of 2,943 Holstein, 1,014 Red Factor, 1,415 Red Holstein, 154 Swiss Fleckvieh and 65 crossbred animals on four farms were examined using a Lely feeding robot. For the analyses, one year before and after the installation of the feeding robot were compared. A mixed linear model was used for the investigations. The model included the fixed effects feeding (with and without robot), lactation, the interaction feeding x lactation, month, a function of day of lactation, as well as farm and animal as random effects and residual effect. The analyses of the milk yield data revealed that the energy-corrected milk yield of first lactation cows with the automatic feeding system was significantly higher by 1.0 kg at 28.2 kg ($p = 0.04$). In the second lactation, the energy-corrected milk yield was significantly higher by 1.4 kg at 32.0 kg ($p = 0.0016$). In all subsequent lactations, the performance with robot was worse than the performance without robot. When feeding with robot, the fat content of the milk was also significantly increased in the first lactation by 0.11 % to 4.40 % ($p = 0.05$). In the second lactation, the effect was no longer significant, the increase in fat content with automatic feeding was 0.09 % ($p = 0.08$). The probability of the occurrence of acetone in the milk was significantly lower in the group with an automatic feeding system (44.8%) than in the group without a feeding robot (53.1%) ($p = 0.008$). The results of the present study indicate that automatic feeding can have positive effects on the milk performance and fat content of young cows.

R4D project - 100 best practices for resilient dairy farming in Europe

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European dairy farmers face major challenges, putting pressure on their resilience. The hurdles they encounter are diverse and ever evolving, ranging from market volatility and environmental pressures to shifting consumer demands and regulatory complexities. A lot of knowledge is already available that could strengthen the resilience of dairy farmers. However, this knowledge does not often reach them. R4D (Resilience for Dairy) project aims to disseminate the most promising solutions to dairy farmers to become more resilient. To achieve this goal, 18 partners created a network enabling exchange of practical and scientific knowledge between European dairy farmers, researchers, and relevant stakeholders. This network assessed practices and solutions that can contribute to resilient dairy farms. This work was completed in several steps: identification of 300 urgent needs at farm level, followed by the collection of 200 potential solutions to answer them, and a multi actor evaluation to rank the proposed solutions according to their score for resilience, readiness for dissemination and acceptability by farmers. Finally, 100 best practices were described and exchanged through leaflets, webinars and cross visits. Improving technical efficiency remains a dominant strategy at farm level, but other aspects like work organization, business management, environmentally friendly production systems, animal health care, and transparent communication with society were also highlighted. Some gaps in knowledge were identified, including solutions for a better communication with society, calling for new research and initiatives to meet the expectations of farmers and consumers.

Session 46

Theatre 2

Resilient future dairy farming systems

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The world faces a wicked problem. To feed the projected global population, annual food production must grow by 30-60% before the middle of the century. But the food system already contributes 20-30% of anthropogenic greenhouse gases, is a significant user of fresh water, is a key contributor to biodiversity loss in some areas of the world and can be a significant contributor of contaminants to fresh and estuarine water bodies. At the same time, the number of people hungry and/or malnourished continues to increase and climate change and significant weather events are reducing food productivity. Furthermore, consumers are demanding more 'nature-friendly' food. Milk is widely recognised as Nature's near perfect food. Neonate mammals have consumed milk as a complete food for more than 200 million years and, with the domestication of lactating cattle around 9,000 years ago, adult humans have consumed it fresh and as preserved products, often as a high proportion of their diet in some nomadic cultures. But dairy farming systems have changed significantly over the last 100-150 years. From predominantly pastoral 'small-holder farms, with surplus pasture harvested in spring as a winter feed, cows milked once- or twice-daily, and diet and lactation length predominantly influenced by available feed supply, to 'mega-dairies', in which animals are housed, milked four to six-times daily, and provided with a consistent ration of conserved forages, grains, and co-products formulated to maximise production. Future dairy systems will have to navigate consumer desires for animal freedoms to exhibit natural behaviours and a more 'nature-friendly' approach to food production, and warmer temperatures and less certain and predictable rainfall patterns. To be resilient, returning a reasonable and consistent income to farmers for their efforts, they will require a system robust to climactic and milk price challenges, a cow able to tolerate warmer temperatures and more variability in feed supply and composition, and an extension/advisory system promoting versatility in operating models.

Towards future sustainable dairy farming systems: An integrated, adapted and circular approach
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Dairy production systems (DPS) are key for European agriculture, providing quality foods, sustaining rural employment, and conserving biodiversity. However, DPS face challenges threatening their viability and resilience, thus necessitating region-specific approaches to ensure its future. Therefore, implementing integrated approaches (economy, society and environment), adopting context-specific strategies, re-connecting crop and livestock production and promoting circularity is key. In this context, the work done in research the projects Eranet "MilKey" and "DairyMix" enabled the identification and design of sustainability and circularity measures that, in combination with an integrated and holistic approach, allow for the adaptation of concepts and strategies to the needs of the sector. This approach provided a better understanding of the effect of emission mitigation options across DPS, facilitating the adoption of tailored and context-specific strategies. At the same time, an innovative framework for assessing synergies and trade-offs across sustainability dimensions was developed. In addition, by making these tools and results available in interactive platforms, informed decision-making is facilitated based on a better understanding of the sector's reality. The combined approach followed, alongside accessible platforms for knowledge dissemination, holds promise in steering European DPS towards sustainability and resilience.

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Theatre 4

Perspectives on further integration of dairy and arable crop production

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More integrated approaches of dairy and arable crop production can be shaped at both farm level and regional level. Such integration offers opportunities to increase ecological, economic and social sustainability of agricultural production. Synergies and trade-offs need to be assessed and well-balanced. At food system level, the balancing of animal and plant production needs to take into account views at circularity, at self-sufficiency levels at different spatial scales, and at healthy human diets. Holistic views on future food systems will be translated into opportunities for further integration of dairy production and arable crop production. These opportunities can be divided in some main categories: 1) the use of leftovers, co-products and crop residues as an input for dairy production; 2) the use of animal manure and processed animal manure fractions as an input for crop production; and 3) land exchange and integrated crop rotation in collaboration between dairy farms and arable farms. These tracks interact and synergies and trade-offs need to be assessed, with soil quality and soil fertility consequences as a key element. In addition, the inclusion of crops for biobased construction materials in crop rotation plans at dairy farms will be showcased. A promising example of how dairy farms can not only contribute to the food system, but to the future biobased non-food system as well. The large diversity of regional and farm characteristics stresses that there will be no generic solutions for integrated crop-dairy systems, However, solutions which are developed in specific regions or in bilateral farm collaborations can provide inspiring building blocks to develop customised concepts to further increase the sustainability and viability of future dairy-crop systems.

Farmer logics – motivations and driving forces influencing a sustainable and resilient Swedish dairy production

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Understanding farmer logics is fundamental to the understanding of how to maintain and strengthen a sustainable and resilient animal production. The intensification and centralisation within the animal production have significantly altered the role of Swedish dairy farmers. The farmers' views of their production as well as driving forces and motives for establishing, developing, and maintaining the production is essential for building a robust primary production. Diversification amongst dairy farms is important for a resilient future food system. Sweden is a vast country with large variations in climate, agricultural land and infrastructure. To ensure a multitude of production systems in terms of herd size, production intensity, and geographic distribution, it is necessary to understand both the on-farm strategies and system level dependencies that lay the foundation for future Swedish dairy production. Previous research has identified different farmer logics unifying primary producers and it could be posited that there is a large variation in drivers, motivators, and logics among Swedish farmers, affecting decision making and development. The aim of this study was to investigate in what way logics apply to dairy farmers in Sweden, to highlight their driving forces and motivations as well as their attitudes on preparedness towards challenges within future food systems. A survey was distributed via e-mail to registered cattle farms in Sweden in April 2023. Respondents agreed on a scale from 1-5 to statements based on results from previous literature. The survey rendered 326 responses from active dairy producers. Regression analysis was used to characterize different types of farmers logics among the responses. This study highlights the importance of understanding farmer logics and the structures behind decisions that may influence the production in different dairy herds. The result can be of value for policy making and regulatory regimes, as well as for advisory services, to facilitate the transformation towards a resilient and sustainable food system.

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Theatre 6

True Cost Accounting for sustainable livestock production

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The sustainability of the food system is at stake, not only climate change but also land use, biodiversity, animal welfare, human health and other social issues are discussed. The production and consumption of food goes along with environmental, social and human side-effects, called externalities, that can be positive but are often negative. Such externalities are not reflected in the price and, thus, insufficiently considered by decision makers. To compare and weight these externalities, True Cost Accounting (TCA) implies to include these externalities in decision making by monetising the externalities and weighing them by the impact on human welfare. TCA uses environmental, social and nutritional LCAs as a basis. The HE project FOODCoST develops a harmonised TCA methodology for food. Livestock products have a higher ecological footprint compared to many other food products like vegetables and fruits. The HE project STEP UP aims to provide a more profound insight in the variation in sustainability performance of European livestock production systems. In FOODCoST, measuring and monetizing the impact of externalities is a first step. This information about sustainability performance can be used to change behaviour. In a second step, businesses, consumers and governments internalise these externalities and change their behaviour to improve sustainability. But how to arrange this? An overview of existing policies divided in administrative and market-based instruments will be given. The option for internalisation of externalities of businesses can be divided in value creation & delivery, value measure and value capture.

Evaluation of the environmental efficiency of innovative dairy cattle housing and management systems in Germany

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While concerns about the climate crisis continue to dominate the public debate on agricultural production, consumers are increasingly concerned about the origin and production conditions of food. For this reason, the germanwide network “InnoRind” was founded, which tests innovative husbandry concepts for cattle, that should go hand in hand with greater consumer acceptance, improved animal welfare and lower environmental impacts. The aim of this sub-project was to analyze how separate areas in the calving area and calf barns for early small group housing affect the level of environmental impacts, with particular consideration of resource use. The study was based on the data of 9 research sites in Germany. Comparative life cycle assessments were calculated for the “baseline“ and the “innovation“ scenario, respectively. As impact categories climate change, freshwater eutrophication, terrestrial acidification and land use were considered. Thereby, the system borders were set from cradle to farm gate and the functional unit was defined as 1 kg energy-corrected milk. Preliminary results showed that the design of the housing system contributes to 9 % of climate change. Comprehensive results on the calves and dairy cows systems will be presented and discussed at the conference. A Monte Carlo analysis will be conducted additionally to quantify data- and system-based uncertainties. The study will provide new insights into potential conflicts of interest between animal welfare and environmental efficiency.

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Theatre 8

Environmental challenges with intensive dairy production

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The successful development of the Dutch Agri & Food sector in the last 70 years by maximizing the production at minimal costs has led to a situation where the Dutch dairy sector is now responsible for a high national contribution to environmental impacts. These are acidification of nature areas (mainly by NH₃) and resulting biodiversity loss, and eutrophication of water bodies (losses of N and P). The global warming issue (emissions of CH₄) has also entered into the discussions. Moreover, a more animal friendly husbandry is demanded by the society. The Ministry of Agriculture and Dairy Organizations stimulate research & development and implementation on dairy farms of an integrated approach to reduce the emissions of nitrogen and greenhouse gasses and stimulate animal welfare. We will present the topics around breeding, nutrition, grassland management, manure management, cattle housing & technology and groups of pilot farms who are working on implementation of the practices and techniques to reach the environmental goals. The expectations for future dairy housing systems is to develop floor types and other techniques that separate feces and urine and maintain walkability, because of the decreased ammonia emissions due to separation, the decreased methane emission due to rapid removal from the barn and the possible creation of different manure products for efficient use of N, P, K and organic matter C. The separation of urine and feces can be performed with a grooved floor or a Cowtoilet that collects the urine before it hits the floor. A group of 150 pilot farms are reducing the protein intake towards 155 gram crude protein per kg dry matter to reduce the ammonia emissions. With feeding and breeding strategies we adapt the animal to the environment. Another strategy is to adapt the environment to the animal. Therefore we study the filtering of ammonia and methane from the air in combination with improving the indoor climate for the animals.

Dear Dairy, is it love or is it over? Australian public attitudes to the dairy industry

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Understanding evolving public views on food production is vital to ensure agricultural industries remain socially sustainable. To explore public attitudes to the dairy industry, a convenience sample of Australian citizens were asked to write their choice of a 'love letter' or 'breakup letter' to the dairy industry. The present study provides results from the 19 letters submitted. Participants varied in age, gender identity, income and frequency of consumption of dairy products. The letters were on average 144 ± 70 words long, and comprised 8 love letters, 6 break-up letters and 5 with a conflicted stance. We undertook inductive thematic analysis of all letters, identifying three main themes: (1) personal relationship with dairy; (2) views about dairy as an industry; and (3) views on dairy products. Support for dairy was mainly communicated through participants' love of dairy products, whilst opposition to dairy largely centered on participants' ethical concerns about farming practices. Some participants were conflicted in their relationship with dairy, struggling to balance their love of the products and their concerns about farming practices. In contrast, participants who conveyed that they had 'broken up' with the dairy industry described an unflinching commitment to their decision. Our findings demonstrate the key role of people's core values in their relationship with dairy. Efforts to identify and address areas of concern that lead to values misalignment with the public may aid in maintaining the social sustainability of the dairy industry.

Session 47

Theatre 1

Effects of hydrolysable tannin metabolites on rumen microbiota and enteric methane production

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Supplementing cattle diets with tannins is a strategy to decrease enteric methane (CH₄) production, mitigating the environmental impact of livestock production. In the present study, a 10-day rumen simulation technique (Rusitec) was used to evaluate the effect of two hydrolysable tannin metabolites, gallic acid (GA) and ellagic acid (EA), and their combination on rumen microbiota. The EA treatments significantly reduced daily CH₄ production by up to 60%. The qPCR showed a reduction of the fibrolytic bacteria *Butyrivibrio fibrisolvens*, *Fibrobacter succinogenes* and *Ruminococcus flavefaciens* as well as an increase of the nitrate-reducing *Selenomonas ruminantium* after EA treatment, thus modulating feed fermentation and in turn methanogenesis. Amplicon sequencing revealed a stronger impact of EA on bacteria than archaea, with reduced bacterial alpha-diversity. Bacterial communities were dominated by *Megasphaera elsdenii* after EA treatments, while archaeal communities were always dominated by the *Methanomethylophilaceae* family. Therefore, EA rather than GA increased the abundance of *M. elsdenii*, likely redirecting H₂ from methanogenesis to other H₂ sinks. However, EA in contrast to GA also induced a decrease of short-chain fatty acids production and nutrient degradation. Overall, EA addition induced stronger effects compared to GA. This underlines the need to assess effects of individual tannin components on rumen processes to develop efficient CH₄ mitigation strategies.

Assessing the potential of six plants from Auvergne region to reduce in vivo methane emissions and nitrogen excretion by dairy cows

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Methane emissions (CH₄) constitute 60% of total greenhouse gas emissions at the farm scale. Our focus was to reduce enteric CH₄ by introducing local plants from Auvergne region in dairy cows' diets. Plants were selected for their ability to reduce CH₄ and nitrogen in vitro. Eight primiparous Holstein dairy cows have been divided into 2 homogenous groups and randomly assigned to 4 diets, following a 4×4 Latin square (LS1, LS2) experimental design with 4 periods of 4 weeks. Diets were fed ad libitum twice daily. The control diet (CTL) was the same for both LS and composed of 70% of fodder and 30% of concentrate (on a DM basis). The 6 other diets were CT supplemented with 90 g DM/cow/day of one of the herbal supplements (HS, 1 to 6): HS 1, 4, 5 in LS1 and HS 2, 3, 6 in LS2. Zootechnical parameters (dry matter intake (DMI) and milk) were measured daily during the 4 weeks. Other measurements included nutrients digestibility and nitrogen balance (3rd week), daily kinetic of enteric CH₄ using the respiration chambers and rumen fermentation parameters (4th week). The statistical analyses were performed using the MIXED procedure of SAS. The 6 HS diets had no significant effect on DMI and milk production, nutrients digestibility, nitrogen balance and rumen fermentation parameters compared to CTL. Only HS2 reduced CH₄ (g/d, g/kg DMI) by -6% (P<0.001) on average compared to CTL and other HS diets. This effect persisted throughout the whole day (hour*HS, P= 0.92). Further investigations (dose, distribution mode) are required in order to optimize the use of these local plants in ruminants' diets.

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Theatre 3

Development, in-vitro and in-vivo assessment of novel feed additives to reduce enteric ruminant methane emissions

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Ruminant livestock contributes significantly to global methane (CH₄) production and its mitigation is of utmost importance, where feed additives represent a cost-effective means of achieving this. Previous research in this project demonstrated that slightly elevating the rumen oxidation reduction potential (ORP) using oxygen-releasing feed additives serves to hinder methanogenesis. This is due to the niche specialisation of methanogens who are typically only active at ORPs below -300 millivolts. In-vitro assessment of these compounds, including calcium peroxide (CaO₂), has demonstrated their effective CH₄ mitigation potential while in-vivo trials revealed no negative impacts on animal performance when supplementing CaO₂ at inclusions of up to 7.25% dry matter intake (DMI). The current work focuses on the impacts of these ORP modulating compounds on the in-vitro and in-vivo rumen microbial communities from the aforementioned trials. DNA and RNA were co-extracted from rumen fluid and amplicon sequencing of the 16S and 18S genes was performed. Shifts in microbial communities were integrated with process data (in-vitro) and animal data (in-vivo) and revealed the extent to which ORP can alter the microbial community. Quantitative PCR will also be performed to determine effects on absolute numbers of rumen fungi, bacteria, protozoa, and methanogens. This work contributes to elucidating the microbial mechanisms underlying the CH₄ reduction that is observed as a consequence of increasing rumen ORP using CaO₂ as a ruminant feed additive.

The effect of an essential oil blend on performance, feed efficiency, methane emission and rumen microbiome in lactating dairy cows

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The aim was to study the effects of a blend of essential oils (EO). The company Agolin SA supplied a product containing 20 % of a blend of EO with the main active compounds: eugenol from clove oil extracts (approx. 25 %), linalool from basil (approx. 15 %) and synthetically produced geraniol (approximately 20 %). Thirty-six Holstein cows were allocated to one of two groups according to parity and DIM and were fed either a standard TMR diet, or the same diet to which was added the EO product containing 0,2 g EO/cow/day. The standard diet was based on 54.4 % of DM from grass-clover silage, 5.9 % from maize silage and rape seed cake and meal, barley, dried beet pulp and minerals. The experiment lasted 10 weeks plus a 1-week pre-period. Methane was measured in GreenFeeders. Dry matter intake (DMI) was measured by RIC feed bins. Rumen fluid was taken with a FLORA® oro-ruminal probe for VFA and microbiome analyses. Rumen microbiome was determined from metagenomic sequencing of DNA from rumen samples and quantitative PCR targeting total bacteria, archaea, fungi and protozoa. There was no effect of EO on DMI. The daily number of visits to the GF was significantly higher for the EO diet. There was no effect of EO on ECM or feed efficiency. There was no significant effect of EO on methane per day, and no effect per kg DMI or per kg ECM. There was no significant effect of EO on any VFA or microbiome parameters. In conclusion, adding 0,2 g of active phytogetic substances per cow per day to a diet based on a high proportion of grass-clover silage had no effect on cow performance, methane emissions, rumen fermentation or microbiome measurements.

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Theatre 5

Does processing of the straw have an effect on methane production from feedlot beef cattle?

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This study assessed the effect of straw processing on methane production from feedlot cattle. Eighteen rumen-cannulated cross bred male beef calves (359±2,7 kg and 250±0,4 days) were fed ad libitum a concentrate and straw in the long form (L) or ground and pelleted (P). Average daily gain (ADG), and intake of both the straw and the concentrate, were recorded through a sixteen-week period. Digestibility was estimated using chromic oxide as marker. Rumen liquid and gas samples were obtained at different times of the day, in two occasions during the experimental period, to determine the rumen fermentation pattern and methane production, respectively. Abundance of total bacteria, methanogens, protozoa and anaerobic fungi was also analysed. All data were analysed using the PROC MIXED of SAS with treatment (and sampling day or sampling time within sampling day, where appropriate) as the fixed effect and animal as random. Initial body weight and age were used as covariates for ADG, and organic matter intake as covariate for digestibility. Rumen fermentation variables (including gas production), and abundance of different microbes were analysed as repeated measures. Straw processing had not a significant effect ($P>0.05$) on ADG, intake of straw, concentrate, dry matter or digestible organic matter, digestibility values, production of CH₄ and CO₂, and abundance of microorganisms in the rumen. Rumen pH was higher ($P=0,042$) for L than for P (6.43 vs. 5.97). Reducing the particle size of straw in cattle fed high-concentrate diets does not seem a suitable strategy to reduce methane production from this type of animals.

Effect of prickly pear (*Opuntia ficus-indica*) in diets on growth performance and methane emissions of yearling male Pedi goats

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The study was conducted to determine the effect of prickly pear (*Opuntia ficus-indica*) in diets on growth performance, and methane emissions of yearling male Pedi goats. A total number of 24 Pedi goats were randomly assigned to 4 treatments in a completely randomized design, replicated six times with 1 goat per replicate. Prickly pear inclusion levels were at 0, 10, 20 or 30%. The experiment was conducted for 28 days (21-day preliminary and 7-day collection periods). The data collected were subjected to analysis of variance using Statistical Analysis System Software. Male Pedi goats fed diets having 10 and 30% Prickly pear meal inclusion levels had higher ($P < 0.05$) DM and OM digestibility values than those fed diets having 0 and 20 % Prickly pear inclusion levels. Prickly pear inclusion level in a diet had no significant effect ($P > 0.05$) on live weight, feed conversion ratios and water intake of goats. Prickly pear inclusion level in a diet had an effect ($p < 0.05$) on methane emissions by Male Pedi goats. Male Pedi goats on a diet having a 0% Prickly pear meal inclusion level had higher ($P < 0.05$) methane emissions than those on a diet composed of 0, 20 and 30% Prickly pear inclusion levels. Although, Prickly pear meal reduced methane emission, optimal reduction in methane emission was not determined. Further validation is required to determine Prickly pear inclusion levels for optimal methane production and emission by goats. It was concluded that indigenous Pedi goat optimal productivity responses to Prickly pear meal inclusion were variable, depending on the production parameter in question.

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Theatre 7

Effects on methane emissions and growth performance in beef cattle supplemented with oil macerate of *Asparagopsis taxiformis*

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The macroalgae *Asparagopsis taxiformis* contains several anti-methanogenic compounds, the strongest being bromoform (CHBr₃). Macerating the macroalgae in vegetable oil yields an oil containing CHBr₃, Bromoil, which can be fed to ruminants. Here, we fed the Bromoil to finishing beef cattle to evaluate its effects on growth and ruminal CH₄ emissions. Twenty yearling crossbred Angus males were randomly allocated to 4 pens. The Control diet consisted of a total mixed ration diet and the supplemented diet was the same diet with added Bromoil supplying 25 mg CHBr₃/kg of feed DM (BrOil). Diets were prepared daily. The animals were kept on trial for 2 months. Live weight and feed intake (DMI) were monitored. Individual methane and carbon dioxide emissions were measured using the Greenfeed® system. Animals were slaughtered in a commercial slaughterhouse and carcass yield was calculated. There were no differences ($P = 0.454$) in average daily gain (ADG) between Control (1.34 ± 0.106 kg) and BrOil (1.23 ± 0.106 kg). The DMI was higher for the Control (10.3 ± 0.14 kg/d) than for BrOil (8.6 ± 0.14 kg/d) ($P < 0.05$). Animals fed BrOil had lower methane emissions (60 ± 16.2 g/d) than those fed Control (180 ± 16.2 g/d) ($P = 0.0001$). The results indicate that the addition of *A. taxiformis* oil extract resulted in a 67% decrease in direct methane emissions without affecting growth rates

Impact of fucosidase on pig growth performance and enteric methane production

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Introduction: Reducing environmental impact while maintaining high growth performance is an urgent challenge in pig production. In-feed fucosidase may cleave fucose from intestinal mucins and release it into the gut lumen, hypothesized to support growth of beneficial gut bacteria and inhibit methanogens. This study therefore aimed to investigate the effect of fucosidase on pig growth performance and enteric methane production. **Materials and Methods:** A total of 240 crossbreed piglets (weaning age 22 days) from 80 sows were included. Three treatment groups (Fuc2, Fuc7, Fuc16) were fed control diets amended with 100 mg fucosidase per kg feed for 2, 7, and 16 weeks, respectively; the control group (Control) received no fucosidase. Average daily feed intake (ADFI) and average daily gain (ADG) were measured. Intestinal digesta were collected on days 10, 43, and 114 for in vitro gas production analysis and quantification of methanogens. Selected pigs (n=80) were placed in respiration chambers (48h) to determine in vivo gas emissions on days 43, 78, and 106. Data were analyzed by mixed effects models and the expected increases from fucosidase amendment were examined with one-sided tests. **Results:** Performance did not differ significantly between control and treated groups ($p>0.05$). The quantification of methanogens did not show significant differences between the fucosidase groups and the Control ($p>0.05$). On day 106, the in vivo methane emission was lower in all three fucosidase groups compared to the Control ($p<0.05$), and the in vitro methane production tended to be lower in the Fuc16 group compared to the Control ($p=0.07$) on day 114. **Conclusion:** The results suggest in-feed fucosidase provision as a potential strategy for mitigating enteric methane emission in pig production.

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Theatre 9

Nitrogen use efficiency from beef fed different protein sources and a forage-based diet

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This study evaluated the effect of three different protein sources (soyabean, SB; brewers' spent grains, BSG; local field beans, BNS) on digestion and nitrogen (N) balance from beef cattle consuming a 70:30 forage to concentrate basal diet, also compared with a 100% pasture-fed beef system (PAS). 16 Holstein × Angus growing steers were divided into four groups, each assigned to one of the experimental diets (SB, BSG, BNS, PAS) with four animals in each group. Every 4 weeks, the animals were moved to digestibility stalls for 4 days for N balance measurements. Dry matter (DM) intake (DMI, kg/day), DM digested (kg/d), N intake (NI, g/day), and N digested (g/d and kg/kg) did not differ between groups ($P>0.05$). PAS steers had higher urinary N output (UNO, $P=0.011$), manure (faeces and urine) N output (MNO, $P=0.039$), UNO/NI ($P<0.001$), and MNO/NI ($P=0.005$). Also, PAS steers had lower digested DM (kg/kg) ($P=0.005$) and N balance ($P=0.025$) than concentrate-fed steers. When concentrate-fed groups were tested alone, SB and BSG had higher N digested ($P<0.001$) and lower faecal N output/NI ($P<0.001$) than BNS. BSG had higher UNO ($P=0.030$) and MNO ($P=0.049$) than BNS and lower ($P=0.002$) DM digested (kg/kg) than SB. Overall, N use efficiency was lower in PAS, likely due to rapid degradation of forage protein in the rumen, and this may have implications to growth rates of PAS steers.

Effect of alpine plant species on in vitro ruminal fermentation characteristics

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The aim of this study was to evaluate in vitro the effect of different plant species widespread on alpine pastures on rumen methane emissions and fermentation parameters. A total of 11 plant species were sampled at the start of the grazing season, coarsely cut, and used as fermentation substrate. An in vitro batch system was used for continuous measurement of gas and methane production and for determining fermentation parameters. A balanced incomplete block design was used, consisting of 11 fermentations where each plant was tested five times. Plants affected all volatile fatty acids analysed after 24h of fermentation. In particular, *Hypericum maculatum* had the highest acetate/propionate ratio, while *Festuca rubra*, *Poa alpina*, *Carum carvi*, and *Lotus corniculatus* had the lowest. Regarding the production of methane, *C. carvi* demonstrated the highest value, while *H. maculatum* had the lowest. Furthermore, *C. carvi* and *Ranunculus acris* have produced more methane per unit of dry matter digested, while *Potentilla erecta* was the species that produced less. Finally, regarding the methane yield kinetics, *C. carvi* and *R. acris* had the same behaviour during fermentation, showing high values for both the maximum methane production rate and the decay in production rate. Instead, the two grasses, along with *H. maculatum* and *P. erecta*, responded in the opposite manner. The variation in methane production among the plants examined was small but significant. In conclusion, *P. erecta* appears to be the plant that best balances methane reduction and nutritional value. In addition, *H. maculatum* likely contains bioactive compounds capable of inhibiting methane production but showed limited digestibility. Study funded by European Union – NextGenerationEU, PNRR (iN-EST, Spoke1, RT2; DM 737/2021).

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Theatre 11

Replacement of total mixed ration with fresh alfalfa on productive performance, nutrient utilization, ruminal environment, and methane emissions of dairy cows

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We evaluated the impact of partially replacing a TMR with fresh alfalfa in lactating cows on intake, digestion, production, and CH₄ emissions. In a crossover-design experiment (29-d experimental period, 21-d adaptation), 12 cows (150 DIM, 545 kg BW, previous lactation: 7000 kg milk), were randomly assigned to T100: 100% TMR, or T60: TMR + fresh alfalfa. The TMR was formulated for 30 kg milk and provided ad libitum in the T100. In the T60, cows were a.m. fed TMR at 60% of the predicted intake and p.m. grazed alfalfa without a limit of quantity. All animals were sampled for rumen environment and CH₄ emissions (SF6 methodology). The intake of DM and nutrients was higher in T100 than in T60 (21 vs. 18 kg DM/d; P<0.001), but no differences were observed in milk production (25 kg/d), milk fat (4.2%), or milk protein (3.4%). Rumen pH was lower (6.2 vs 6.43; P=0.01) and propionic tended to be higher (20.6 vs. 19.8 %; P=0.06) in T100 than in T60, due to the higher starch content. Butyric was lower in T100 (10.5 vs 11.1%, P=0.03), probably due to the higher sugar content of fresh herbage. The cows in T100 emitted less CH₄ related to intake (25 vs. 29 g CH₄/kg DM, P=0.03; 48 vs. 54 g CH₄/kg NDF, P=0.04), but there were no differences in CH₄ emitted per day (523 g CH₄/d), per kg of milk (22 g CH₄/kg milk), milk protein (628 g CH₄/kg protein) and milk fat (518 g CH₄/kg fat). We concluded that the partial substitution of TMR by fresh alfalfa in dairy cows led to a rumen environment similar to that of cows fed only TMR, without increasing CH₄ emissions or negatively affecting milk production.

Enteric methane and digestibility: are these phenotypes correlated in dairy cows?

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Selection of cows with low methane (CH₄) emission would reduce emission from the dairy sector. However, if low CH₄ emitting cows are less efficient in digesting fibre, such selection for low CH₄ emitting cows could result in selection towards cows that are less efficient in fibre digestion, offsetting the benefits of the reduced emission. Cow estimates were obtained from a linear mixed model by removing the CH₄ reducing dietary treatment effects from three Latin square trials. Cow estimates were obtained for DMI (kg/d), energy corrected milk yield (ECM; kg/d), CH₄ production, yield, and intensity (g CH₄/d, g CH₄/kg of DMI, g CH₄/kg of ECM, respectively), and digestibility of dry matter, organic matter, and neutral detergent fibre (DMD, OMD, NDFD, respectively; %). The estimated mean intercept and between-cow variation (mean ± SD) for DMD, OMD and NDFD were 72.2 ± 1.1, 73.8 ± 1.1, and 60.4 ± 1.9 %, respectively. The mean intercept and between-cow variation for CH₄ production, yield and intensity were 362 ± 42 g/d, 16.9 ± 1.6 g/kg of DMI, and 9.93 ± 1.37 g/kg of ECM. For DMI and ECM yield the mean intercept and between-cow variation were 22.3 ± 2.2 kg/d and 37.0 ± 5.1 kg/d, respectively. When correlating cow estimates for enteric the OMD was found to be positively correlated to CH₄ yield and intensity (r = 0.25 and 0.37, respectively; P ≤ 0.001), and so was NDFD (r = 0.41 and 0.49, respectively; P ≤ 0.001). The OMD and NDFD were negatively correlated with ECM yield (r = -0.28 and -0.29, respectively; P < 0.001). The efficient digestibility phenotype is positively correlated with CH₄ emission in terms of CH₄ yield and intensity, with the possible risk of reducing OM and NDF digestibility when merely selecting cows for low CH₄ yield and intensity.

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Poster 13

Herd-level greenhouse gas emissions of dairy rations optimized for reduced carbon footprint

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Improving the environmental sustainability of dairy production through dietary interventions requires assessment at different hierarchical levels for the presence of trade-offs. This study estimated the greenhouse gas emissions (GHG) of 130 high-yielding Swedish Holstein cows and recruitment animals. Data were collected from an on-farm animal experiment conducted in 2022 with direct measurements of enteric methane (CH₄) using the Green-Feed system. The results were transformed to fit calculations on an annual basis. The NorFor feed evaluation system was used to formulate rations based on grass-clover silage and three different concentrate mixes: standard commercial (CON), by-product-based (BYP) considering aspects of feed-food competition, or home-grown crops (HOM). Annual farm gate deliveries were set at 1235 t. energy corrected milk (ECM) and 31.3 t. of live weight (slaughtered cows, male and female offspring) and required 1497, 1569, and 1524 t. of feed dry matter for CON, BYP and HOM respectively. Herd total CH₄ yield was: CON: 28.5 Mg/y, BYP: 28.0 Mg/y and HOM: 28.9 Mg/y. Feed GHG expressed as carbon dioxide equivalents (CO₂e) were: CON: 653.4 Mg/y, BYP: 570 Mg/y, HOM: 606.7 Mg/y. Emission intensity, based on the sum of CH₄ and feed carbon footprint and allocated according to the International Dairy Federation guidelines, was: 1022, 955, and 998 g CO₂e/kg ECM for CON, BYP, and HOM, respectively. Our results indicate that implementation of BYP and HOM diets on a herd level could reduce the emission intensity by 6.6% and 2.4% compared to CON. This work was carried out within the collaborative centre SustAnimal.

How much of the between-cow variation in enteric methane production can performance and digestibility measures explain

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A previous study found that dry matter intake (DMI) alone explains a similar proportion of between-cow variation in methane production (CH₄; g/d) as performance measures when grouped together. The aim of this study was to test if inclusion of digestibility measures along with performance measures increases the proportion accounted for of between-cow variation in CH₄ production. The cow estimates were obtained from a linear mixed model by removing the CH₄ reducing dietary treatment effects from three Latin square trials. The cow estimates of the performance measures DMI, energy corrected milk yield, milk fat, protein, and lactose proportions, and metabolic body weight, together with cow estimates of digestibility of dry matter, organic matter, and neutral detergent fiber, were scaled and used as explanatory variables in a principal component regression model with cow estimates for CH₄ production as dependent variable. Compared to DMI alone, the use of grouped performance measures increased R² from 0.24 to 0.37 (+50 %), and additional inclusion of digestibility measures further increased R² to 0.51 (+100 %). This was a bigger increase in explained between-cow variance for CH₄ production compared to a previous study, which found a maximum of 20 % increase in R² when using grouped performance measures compared to DMI alone. The explained variance for DMI alone was lower compared to the previous study, probably due to the reduced number of individuals used in the present analysis, as digestibility data were not available for all individuals in the previous study. Including digestibility measures gave the highest explained variance for CH₄ production than performance measures alone, but 50 % of the variance remained unexplained.

Development of an Effective Microbial Agent Utilizing Beneficial Microorganisms for Odor Reduction and its On-Site Evaluation in Pig Farms

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Microorganisms Suitable for Feed Additive Guidelines *Bacillus subtilis* strain was used for feed and slurry additives in combination with *L. plantarum* LP11 and *Bacillus siamensis* in the swine farm-scale trial for 77 days. The number of viable live microorganisms contained in the probiotics supplied to fattening pigs in feed, used for cleaning the pig houses and that used in in slurry treatment. The infeed probiotic contained 6.34 log₁₀ cfu/g *Bacillus*, 6.78 log₁₀ cfu/g Lactic acid bacteria and 6.62 log₁₀ cfu/g of yeast. There were no Coliforms detected in this product. Fattening pigs were provided with a daily ration of feed containing at least 10⁶ cfu/g of the probiotics and at least 10⁶ cfu/g of *Bacillus* solution was applied inside the pig houses and the slurry pit. Among the volatile fatty acid substances, odor contribution for valeric acid was reduced by 34.55% compared to before application of the probiotics. Styrene, a volatile organic compound and Hydrogen sulfide, a substance with high odor contribution in the pig compost facilities were reduced by 91% and 99.95% respectively. Additionally, p-Cresol belonging to phenols and skatole belonging to Indoles were reduced by 46.98% and 72.46% respectively. The results thus show that odor contribution was improved by about 47% from 2,169 before the application of microorganisms to 1,151 after the application of microorganisms. These results provided a comprehensive understanding of odor-reducing and probiotic characteristics of the *Bacillus* strains and their potential applications to improve the swine farm environment.

Effect of a molasses based liquid feed on diet digestibility, volatile fatty acid production and methane emissions in lactating dairy cows

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Molasses based liquid feeds (MBLF) can significantly improve fibre digestion, nitrogen retention, volatile fatty acid (VFA) production and rumen pH thus increasing forage utilisation efficiency and reducing protein supplementation requirement. Limited research suggests sucrose feeding favours butyrate production and has neutral impact on methanogenic populations. The hypothesis is that methane production should be neutral or lower for cows with higher sugar diets. An 8-week crossover design study with 28 lactating dairy cows consisting of two periods and 2 diet treatments, including a control diet and treatment where 1.4 kg DM of concentrate was replaced with the MBLF 'Regumix' on a CP equivalent basis. Measurements included dry matter intake (DMI), milk yield (MY) and composition, rumen pH and VFA. Four cows underwent additional measurements of total tract digestibility and CH₄ emission within respiration chambers. Data was statistically analysed using the Mixed procedure of SAS® and a model testing fixed effects of treatment and the random effects of cow and period. DMI, MY and N balance were unaffected by treatment. Milk fat concentration increased ($P < 0.05$) in cows receiving MBLF. Methane production and intensity in relation to DMI and MY was similar for both treatments. A reduction in milk nitrogen g/day ($P < 0.05$) and increase in urinary nitrogen and VFA concentration ($P < 0.05$) was observed in the MBLF treatment. The inclusion of MBLF in the diet as a replacement for concentrate feed maintained levels of production, increased milk fat concentration VFA production without negatively affecting diet digestibility, nitrogen balance or methane production.

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Poster 17

Dose effects of feed supplemented calcium peroxide as a methane inhibitor on feed intake and digestibility in beef cattle

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Reducing enteric methane (CH₄) production is of global importance and feed additives represent a valuable mitigation tool. The rumen oxidation reduction potential (ORP) parameter influences CH₄ production rates, as methanogens are typically active only at ORPs below -300 millivolts. Previous in-vitro work demonstrated successful CH₄ mitigation when supplementing with various oxygen-releasing compounds, including calcium peroxide (CaO₂), which modulated rumen ORP. This research evaluated the in-vivo effects of various doses of CaO₂ on feed intake and digestibility for the first time in beef cattle. Nine cannulated Aberdeen Angus × Friesian steers (739 ± 67 kg BW) were used in an 84-day Latin Square design trial (n=9). Varying CaO₂ inclusion rates were assessed during once-a-day and twice-a-day feeding, while in-vivo ORP, in-sacco diet digestibility, palatability, rumen microbiome, pH, ammonia, and volatile fatty acids concentrations were all monitored. Preliminary CH₄ emission indications were obtained via a portable CH₄ analyser. No negative impacts of inclusion rates of CaO₂ up to 7.25% of dry matter intake (DMI), with positive indications of reduced enteric CH₄ emissions. CaO₂ was deemed suitable to progress to larger-scale animal trials and to be incorporated into a feed pellet format, with potential to be feasible in both intensive and pasture-based production systems.

Development of an odor-reducing probiotic *Bacillus* strains isolated from swine feces and Korean fermented soybean pastes

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This study was conducted to develop odor-reducing probiotic *Bacillus* strains for potential use as feed and slurry additives. The 115 strains were isolated from swine feces and Korean fermented soybean pastes, and six *Bacillus* strains were selected based on the evaluation of probiotic characteristics and proteolytic activities. The potential strains were *B. siamensis* G1-1 and G4-M9, *B. amyloliquefaciens* G1-15, *B. pumilus* G3-11, *B. subtilis* G4-N7, and *B. subtilis* subsp. *Inaquosorum* G4-N7. All strains exhibited odor reduction activity for dimethyl sulfide gas. The strains G1-15, G3-11, and G4-N7 exhibited odor reduction activity for ammonia. The G4-N7 strain exhibited the highest odor reduction activities in both odorous gases. These results provided a comprehensive understanding of odor-reducing and probiotic characteristics of the *Bacillus* strains and their potential applications to improve the swine farm environment. To isolate beneficial microorganisms and confirm the suitability of microbial probiotics, bacterial isolates were analyzed using 16S rRNA gene amplification and sequencing. For accurate evaluation of stability, strains were selected based on acid and bile salt tolerance and heat tolerance. The selected strains underwent proteolytic activity tests and antibacterial analyses. Through these processes, two strains, *Bacillus subtilis* subsp. (G4-N7) and *Bacillus siamensis* (G4-M9), were chosen based on their odor reduction potential measured by *in vitro* slurry fermentation and their capability for pig slurry nitrogen decomposition capacity.

The updated FEEDAP Guidance on the assessment of efficacy of feed additives: additives which favourably affect the environmental consequences of animal production

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Within the current climate emergency, agriculture is estimated to account for 10.3% of total greenhouse gas emissions in the European Union and nearly 70% of these emissions are derived from the animal production sector. While transitioning towards a more sustainable food production system, the European Commission aims to promote the development of feed additives that could mitigate the effect of animal production on the environment (e.g., reduced N/P excretion, lower methane emissions). From the European regulatory point of view, these additives are categorised as ‘zootechnical additives’. The authorisation process requires demonstrating efficacy through *in vivo* animal trials to support the claimed effect of the additive on the target animal species, and the assessment of these efficacy trials is the responsibility of the EFSA FEEDAP Panel. Currently, very few applications are submitted for authorisation within the functional group of “substances which favourably affect the environment”, thereby suggesting great possibilities for research and development in this field. In this scenario, the EFSA FEEDAP Panel took the opportunity to update the Guidance on the assessment of efficacy of feed additives to (i) develop further the potential direct and indirect beneficial effects on the environment expected from this type of additives and (ii) to clarify the requirements of the studies submitted to support the efficacy of the new products placed on the market.

A review of dairy production performance on different methane reducing dietary treatments: 3-NOP, nitrate, linseed and rapeseed

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In the past decades, methane-reducing rations and feed additives have been developed, with some currently governmentally subsidized in certain regions (e.g. Flanders (Belgium)). However, voluntary implementation by dairy farmers is progressing slowly, due to cost and scepticism about effects on animal performance. To address this, a paper review was conducted to assess the effects of several methane reducing feed measures on performance. Four methane-reducing dietary treatments were examined: 3-NOP, nitrate (NIT), extruded linseed (EL), and rapeseed (R). A total of 36 papers with 75 in vivo treatments were withheld. Regression analyses were performed for effects on different performance parameters (milk production and composition, dry matter intake (DMI),...), as a function of dose. Additionally, intersecting points were identified for the doses currently subsidized in Flanders, being 60ppm (3-NOP), 10g/kg DM (NIT), 400g/day (EL), and 350g/day (R). Results revealed strong variability between papers for most parameters. Milk production varied between -11% and +10% at the same dose compared to control groups. Effects appeared to be dependent on the baseline ration in which the feed/additive was administered, especially for EL and R. For NIT, no effects on performance were found. For 3-NOP, a trend towards lower DMI was found for higher doses, but the intersection at the 60 ppm dose showed only a numeric 1.6% reduction. In conclusion, for the doses subsidized in Flanders, no consistent effects on production parameters were observed, neither positively nor negatively. Further investigation into the complex interplay of feed additives with the baseline diet is warranted.

Effects of Anavrin® on dry matter intake, ruminal environment, and methane emissions of finishing steers

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Twelve finishing steers (16.8 ± 2.0 months old and weighing 362 ± 24.7 kg), were randomly assigned to a replicated 2×2 cross-over design with 41-d periods T1) CONTROL: total mixed ration (TMR containing 12:88 forage:concentrate ratio on a DM basis; 80.0 % DM, 11.8 % CP, 18.3 % NDF) without additive, T2) ANAVRIN: the same TMR supplemented with 0.35 g of Anavrin® per 100 kg of body weight (BW). All animals were rumen sampled by 8 mm rumen catheters (16 samples over 24-h) to determine pH and volatile fatty acids (VFA). Enteric CH₄ emissions were quantified through the SF6 methodology, and individual dry matter intake (DMI) was calculated for each steer as the difference between the TMR offered and rejected. Data were analyzed using analysis of variance and the MIXED procedure of SAS. Treatments did not affect DMI (14 kg/d), but ANAVRIN produced lower daily CH₄ emissions than CONTROL (156 vs. 189 g/d; P=0.04), and reduced CH₄ yield per kg of DMI (11.4 vs. 13.5 g CH₄/kg DMI; P=0.043) and yield per kg of metabolic weight (1.47 vs 1.77 g CH₄/kg BW^{0.75}; P=0.035). No differences were observed in pH (5.9), total VFA production (87.6 mM), or the proportions of acetate, propionate, and butyrate (50.5%, 35.6%, and 13.9% respectively; P>0.05). Our results show a positive effect of Anavrin® on the reduction of methane emissions and suggest higher efficiency of energy use in animals receiving the additive. Further investigation into the effects on rumen microbiology is ongoing for a more comprehensive understanding of the observed differences.

Substitution of soybean meal with rapeseed meal decreases the ratio of urinary to fecal nitrogen excretion in growing pigs

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Understanding the influence of protein and fibre sources, as alternatives to soybean meal (SBM) and/or corn, on nitrogen (N) balance in pigs is crucial for optimal performance, economic efficiency, and mitigating environmental impacts associated with N excretion. Four experimental diets were formulated: SB, based on SBM (16%) and corn; RS and SF, with 100% replacement of SBM by rapeseed meal (RSM) and sunflower meal (SFM), respectively; and RS-SB, as RS with additional 35% partial replacement of corn by sugar beet. Diets were fed to 12 pigs over 4 periods in a 4×4 Latin square design (3 animals per treatment and period). Diets included 50 mg/kg of ytterbium oxide as indigestible marker. After 7 days of adaptation, daily urine and feces samples were collected over 2 days. N intake (iN), excreted in feces (fN) and urine (uN), totally excreted (eN), and retained (rN) were determined for diets (/kg diet) and ingredients (/kg) following the ingredient substitution method. Data were analysed using the 'glmer()' function in R Studio. The N contents in SO, RS, SF, and RS-SB were 24, 20.5, 21.1, and 19.5 g/kg, respectively. Results showed that eN:iN ratio was greater (P<0.001) in SF (0.37) than in SB, RS, and RS-SB (0.29-0.31) and that uN:eN ratio was higher (P<0.001) in SB (0.64) and SF (0.62) than in RS (0.49) and RS-SB (0.48). Regarding the ingredients, the crude protein concentrations in SBM, RSM, and SFM were 46.2, 32.4, and 37%, respectively. The rN:iN ratio was higher (P<0.001) in SBM (0.62) than in RSM (0.54). The incorporation of RSM promoted a change on the N profile in manure, as fN:uN ratio was higher (P<0.001) in RSM (1.61) than in SBM (0.60).

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Poster 23

Reduction of CH₄ production in ruminants using encapsulated feed additives in vitro

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Organic compounds are one of the most promising additives to mitigate CH₄ emissions from ruminants, but their use in the field can be challenging due to their volatile properties. One strategy is to encapsulate such compounds. This study aimed to evaluate the effect of 7 encapsulated compounds on ruminal fermentation in vitro. The compounds (A1 to A7) produced by SILVA® were tested using two different diets (D1=50:50, D2=15:85 – forage: concentrate ratio). The doses tested were (0.25, 0.5, 1, and 2%). 0.5 g of each diet was placed into bottles containing 50 mL of buffered ruminal fluid and the additives. After 24 h, dry matter digestibility (DMD), volatile fatty acids (VFA), and CH₄ production were assessed. Only ruminal fluid was used as control. Results showed that with D1, additives decreased not only decreased CH₄ production but also DMD and VFA production. When incubating the additives with D2, CH₄ was significantly reduced by 5 out of 7 additives without detrimental effects in other parameters, suggesting their potential application in the feed industry. In conclusion, 5 out of 7 encapsulated feed additives were effective when used with a 15:85 forage: concentrate diet, even at the lowest concentration. However, when incubating a diet richer in forage, the same compounds adversely affected other parameters, suggesting possible negative implications for the animals.

Do cows visit the GreenFeeder randomly?

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Methane (CH₄) emission from GreenFeeders (GF) must be based on at least 20 measurements with a duration of 2 to 5 minutes, whereas measurements from respiration chambers (RC) lasts for at least 24 hours. The CH₄ emission from cows has a diurnal pattern related to feed intake. The assumption behind the GF system is that cows' visits randomly over the 24 hours. The aim of this study was to investigate if cows visit the GF randomly or the visits were associated with meals ingested outside the GF. Data for the analysis were from an experiment testing two different diets. Information on visits and feed intake were from the Roughage Intake Control (RIC) bins. The 35 cows were allocated to 3 GF units. Time from end of a meal in the RIC system until the first visit in the GF system, and from the end of GF visit until the start of new in RIC was calculated. In total 5722 GF visits were included. The average CH₄ emission was 611±159 L/day, the average size of the meals in RIC bins before GF visits was 1.6±0.9 kg dry matter. The cows visited the GF unit on average 71±91 min after the end of a RIC visit, and 32 % of the visits were performed within the first 2 minutes after the cows left the RIC bins. The cows visited the RIC bins 266±177 min after their last GF visit, showing that cows go to the GF closer to the previous meal than to the next meal, which may influence the methane measurement in the GF. There was a significant effect of diet on the number of visits to the GF, and for the diet with the most visits there was a significantly shorter time from a RIC visit to a GF visit. There was no difference between diets for the CH₄ emission, size of meals in RIC bins and time between end of a RIC meal until a GF visit. In conclusion, visits in the GF unit are not random but connected to the feeding behavior of the cows.

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Poster 25

Effect of pastures with rich tannin legumes on ruminal environment and methane emissions of beef cattle

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The aim of this study was to evaluate the ruminal environment and methane emissions of beef suckling cows grazing native improved pastures with a rich tannin legume (*Lotus subbiflorus*). Fifty-six Aberdeen Angus×Hereford multiparous cows (LW 497±6.8 kg; BCS 4.05±0.04 units) were assigned to each of two treatments on Day 22±0.82 postpartum: (i) native pasture (NP; n=28); (ii) native pastures improved with *Lotus subbiflorus* (improved pastures, IP, n=28). The forage allowance was 4.7 kg DM/kg LW in both treatments. The proportion of legume in IP was 27% with 3.6g/100 g DM of condensed tannins. Methane production was measured for 49 days using the GreenFeed system (GF; C-Lock Inc., USA) and less animals were effectively measured (n=11 and n=10 for NP and IP respectively). These animals were rumen sampled by oesophageal intubation to determine pH, volatile fatty acids (VFA) and NH₃. The data were analyzed with the MIXED procedure of SAS (9.0; USA) with a model that included the fix effect of the diet. Cows in IP presented lower (P<0,05) ruminal Acetate concentrations and higher (P<0.01) NH₃ content respect to cows in NP. VFA, Propionate and Butyrate concentrations and ruminal pH did not differ between treatments. Methane production was higher (P<0.001) in cows in IP than in NP (359 vs 296 g/a/d). The intensity of methane emissions was lower in IP respect to NP (171 vs 220 gr/kg LW cow + calf; P=0.05). Native pastures improved with *Lotus subbiflorus* reduced ruminal acetate concentration and intensity of emissions of beef cattle cows, being a possible nutritional strategy to reduce methane emissions in rangeland systems.

Effects of biochar in lactating dairy cows diet

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The feeding of biochar (BC) in livestock as a carbon dioxide sink or carbon storage is controversially discussed. BC has been suggested to improve animal health and performance and reduce enteric methane emissions. However, there is a lack of reliable data for its methane mitigating effect. Therefore, in the present study we investigated the addition of BC to a balanced ration of lactating dairy cows. For the study eight lactating Holstein dairy cows received a basal ration with and without 1% BC on a dry matter basis and were fed over two 36-day periods in a crossover design the respective diet. During the last week of each period, the cows were kept in tie stalls where milk yield and quality, feed intake and nutrient digestibility were quantified. Additionally, the animals were kept for the last two days of the feeding period in respiration chambers for gas emission measurements. The results showed that BC feeding had no significant effect on milk yield, nutrient digestibility and enteric methane emissions compared to the control diet without the addition of BC. Furthermore, feed intake was not affected by the addition of BC and no health problems of the animals in response to the treatment were observed. The results indicate that BC is not an effective feed additive for methane mitigation in dairy cows. However, the beneficial effects of feeding BC to animals suffering from health problems cannot be excluded.

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Poster 27

Metabolomic Approach for the Quality Assessment of *Ulva australis* as a Promising Feed Supplement for Reduction of Methane Emission in LivestockH. Hyeon¹, E. B. Jang¹, M. Kwon¹, D. J. Oh¹, E. Kim², W. J. Yoon¹¹ Biodiversity Research Institute, Clean Bio Business Division, Jeju Technopark, 63608 Jeju, South Korea, ² IANS Co., LTD., 31090 Chungcheongnam-do, South Korea

Ulva australis is a marine macroalgae distributed along coasts of Jeju Island, Korea, regarded as an undesirable green tide which causes serious adverse effects on coastal tidal and subtidal ecosystems. However, *U. australis* has been recently found to reduce methane emission in livestock rumen and to exhibit anti-obesity and anti-diabetic activities. To develop *U. australis* as forage supplement of rumen livestock, metabolic profile of *U. australis* extracts was conducted by gas chromatography-mass spectrometry (GC-MS), gas chromatography-flame ionization detector (GC-FID), and high-performance liquid chromatography-diode array detector (HPLC-DAD). A total of eight low-molecular sugars were identified by GC-MS, including glycerol, glucose, fructose, mannitol, mannose, inositol, galactose and sucrose. Through GC-FID analysis, total eleven fatty acids were quantified; these were lauric acid, myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, arachidic acid, behenic acid and erucic acid. Moreover, natural pigments (chlorophylls and carotenoids) present in *U. australis* were analyzed using HPLC. The examined primary and specialized metabolites data were subsequently applied to multivariate analysis such as principal component analysis (PCA) and hierarchical cluster analysis (HCA). Our results seemed to be ideal for the quality assessment of *U. australis* in response to harvest times and useful for developing *U. australis* resource as a promising feed supplement for reduction of methane production in livestock.

Exploring Microbial Community Dynamics in Cow Gastric Juice with Seaweed-Enhanced Feed: Implications for Methane Reduction Strategies

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Methanogenesis is central to anaerobic digestion processes in ruminant stomach as methane generation. The utilization of seaweed-supplemented feed in cattle has been demonstrated to mitigate methane emissions, yet the underlying pathways and microbial interactions remain poorly elucidated. This study aimed to investigate shifts in the microbial community within bovine gastric juice following the introduction of seaweed feed, specifically targeting microorganisms involved in methane oxidation and symbiotic relationships under anaerobic conditions. Through comprehensive analysis, we observed distinct fluctuations in microbial composition, with certain taxa declining while others proliferated over time. Notably, alterations in the microbial community structure corresponded to consistent changes in the relative abundance of two to three distinct communities, highlighting coordinated shifts rather than isolated events. These findings suggest a complex interplay among microorganisms, wherein symbiotic organisms may influence the activity of methane-oxidizing microbes within the anaerobic cow stomach. This study underscores the potential for indirect methane mitigation by modulating the activity of symbiotic organisms, presenting a promising avenue for reducing methane emissions in livestock systems. Such insights contribute to the development of sustainable agricultural practices aimed at mitigating greenhouse gas emissions and promoting environmental stewardship.

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Poster 29

Evaluating the potential of 3-nitrooxypropanol to reduce enteric methane emissions of autumn-calving cows

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Enteric methane represents 62.5% of Irish agricultural green-house-gas emissions and given targets of reducing agricultural emissions by 25% by 2030, assessing means to reduce enteric methane emissions is vital. In Ireland, winter-milk systems consist of autumn-calving cows that are primarily pasture-based but are housed for a period in early lactation due to poor weather or pasture availability. This provides an opportunity to include methane mitigating feed additives in their diets. The objective of this study was to evaluate the effect of 3-nitrooxypropanol (3-NOP) on enteric methane production in an Irish winter-milk scenario. Eighty Holstein-Friesian cows were blocked based on pre-experimental milk production, methane emissions and parity and then randomly assigned to one of two dietary treatments. The treatments consisted of cows offered a diet containing 78 mg of 3-NOP/kg of DM (TRT) or a control diet containing no 3-NOP (CON). Diets consisted of 32% grass silage, 32% maize silage and 36% concentrate and were fed to achieve 5-10% refusals. Enteric methane emissions were measured using two GreenFeed units across a 6-wk period. There was no effect of diet on milk yield ($P=0.019$). However, cows fed TRT had greater milk protein concentration and milk solids (MS) yield when compared with CON (3.55% and 2.48 kg/d vs 3.50% and 2.42 kg/d, respectively; $P<0.05$). Cows fed TRT had 26% lower methane production and 28% lower methane intensity when compared with CON (335 g/d and 136 g/kg MS vs 452 g/d and 190 g/kg MS, respectively; $P<0.001$). Overall, the results are promising for methane mitigation during housed periods; however, assessment of other strategies will be needed for Irish pasture-based dairy systems.

Effects of oxidising methane inhibitors on emissions, fermentation patterns, and rumen microbial communities in vitro using the rumen simulation technique

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Enteric methane (CH₄) is responsible for 6% of total global greenhouse gas (GHG) emissions and is the primary source of GHG within the agriculture sector. This study assessed oxidising compounds as antimethanogenic agents with future potential to be included in ruminant feeds. Low concentrations of oxidising agents, namely urea hydrogen peroxide (UHP) with and without potassium iodide (KI), and magnesium peroxide (MgO₂), were investigated for their effects on CH₄ production, total gas production (TGP), volatile fatty acids (VFA), nutrient disappearance, and the rumen microbial community in vitro, using the rumen simulation technique. The rumen microbial community was investigated using 16S and 18S rRNA profiling on the Illumina MiSeq platform, alongside quantitative PCR of total bacteria, fungi, protozoa and methanogens. Five treatments were tested (Control, 0.5X UHP+KI, 0.5X UHP, 0.25X UHP, and 0.12X MgO₂), and the in vitro diet consisted of 50:50 grass silage:concentrate on a dry matter basis. All treatments reduced TGP and CH₄ parameters. Maximum reductions were achieved by 0.5X UHP+KI, reducing TGP by 42% (p = 0.091), CH₄ by 53% (p = 0.002), and CH₄ mmol/g digestible organic matter (DOM) by 73% (p = 0.011). The degradation of neutral detergent fibre in the silage substrate was reduced by higher concentrations of UHP. UHP reduced molar proportions of acetate and butyrate while increasing propionate and valerate. Quantitative analysis demonstrated that MgO₂ had the least effect on numbers of key microbial taxa, whereas UHP notably reduced numbers of protozoa and fungi. Within the eukaryotic microbial community, all treatments except MgO₂ tended to reduce the relative abundance of ciliate protozoa. These data offer novel insights into how these highly effective methanogenic inhibitors reduce CH₄ by altering microbial community dynamics.

Session 48

Theatre 1

Deriving genotypes of non-genotyped horses

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Breeders of sport horses frequently use semen from competing studbooks. These open populations pose challenges when it comes to parental verification. KWPN genotypes all live-born foals using Illumina's Equine 70k bead chip for purposes such as pedigree verification and breeding value estimation. However, due to the use of competitor semen, not all stallion genotypes are accessible for parentage verification. If a stallion has enough genotyped offspring, especially with genotyped mates, it should be possible to derive its genotype. The research question of this study was: What number of genotyped offspring (NOFF), given the percentage of genotyped mates (%GM), is needed to obtain the minimum required accuracy of a derived genotype. For this study, genotypes from sires were removed, derived using FImpute and compared with the observed ones. 75 stallions have been selected to represent all combinations of NOFF \bar{x} = 61 (ranging from 4 to 631) and %GM \bar{x} = 32 (ranging from 0 to 100%) as good as possible. Genotypes were only used if they exceeded a call rate of 95%. The sex chromosomes and SNPs with a MAF below 1% were removed. The traditional pedigree contained 3 generations. The trait of interest was defined as the percentage of genotypes that differed between the observed and the imputed ones (%DIFF). A multiple regression analysis ($r^2 = 0.89$) established the prediction equation for %DIFF, using the reciprocal of NOFF and its interaction with %GM as independent variables (p < 0.0001 for both). At 50 NOFF and 100 %GM or at 127 NOFF and 0 %GM the theoretical asymptote of 0% was reached. Imputation corrects for genotyping errors, theoretically resulting in higher accuracy for derived genotypes. Therefore, it is questionable whether 0 %DIFF is the preferred threshold. Accepting a 2% threshold would require 10 NOFF if all mates were genotyped and 25 without mate genotyping.

Single-step GWAS of factors for young horse test traits in Swedish Warmblood horses

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Young horse test scores for conformation, gait and free-jumping traits function as early indicators of show jumping and dressage talent in Swedish Warmblood horses (SWB). This study aimed to investigate possible candidate genes/regions for young horse test traits. Factor analysis was done for eight evaluated traits together with height at withers for 20,814 SWB, and for 50 linearly scored traits with height at withers for 4,782 (of the 20,814) SWB, using the Psych package in R. Among those horses, 380 were genotyped using a 670K SNP array. All genotyped horses had evaluated trait scores and 379 were linearly scored. Single-step (ss) GWAS was done using the BLUPF90 suite of programs for four factors for evaluated traits and 13 factors for linearly scored traits. Significant SNPs associated with three factors related to size were located on ECA3 within or nearby the LCORL/NCAPG region. Significant SNPs were also detected for two factors for evaluated traits representing conformation and jumping, and four factors for linearly scored traits related to body length, neck conformation, walk, and trot (hindlegs), respectively. Among nearby genes, Calcium/Calmodulin-Dependent Protein Kinase Type 1D (CAMK1D) for the factor for linearly scored traits related to neck conformation, and GLI Family Zinc Finger 2 (GLI2) for the factor for evaluated jumping traits, seemed most relevant. For these, top associated SNPs were within the genes, and the known gene functions may be related to the phenotypes. In conclusion, ssGWAS is beneficial to detect plausible candidate genes/regions for desired traits in warmblood horses.

Session 48

Theatre 3

Development and Genetic Architecture of White Markings in Heavy Warmblood Horses

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White markings (WM) are widespread in domestic horses and are physiologically of high interest because of their early point of origin in embryonic development. Extensive WM or spotting is negatively associated with vitality and health. We aimed to investigate the development of WM in the Saxon-Thuringian Heavy Warmblood (STHW) horse and to gain insights into the heritability and genetic architecture of this trait. A quantitative WM-score was derived from the written WM description for STHW born between 2000 and 2023 (N = 9,519). WM on the head, legs as well as body were scored according to their distribution and summed up into scores for the entire animal (TS), head (HS) and legs (LS). Pedigree information was available with a mean pedigree depths of 5.83 full generations. SNP genotypes (Illumina Equine80select) were available for 943 horses, out of which 761 were phenotyped. GWAS with leave-one-chromosome-out approach were performed in GCTA and BLUPF90. No significant changes were observed over the birth years for the TS, HS and LS phenotypes. While there was synchronicity between the leg pairs, a slight but significant lateral asymmetry ($p \leq 0.05$) was observed. The heritabilities for WM ranged between 0.15 (muzzle, s.e.=0.02) and 0.56 (forehead, s.e.=0.025), were moderate for the single leg scores (0.32-0.42, s.e.<0.026) and were highest for TS, HS and LS ($h^2 = 0.72, 0.71, 0.61, s.e.<0.024$). High genetic correlations ($r_g > 0.9$) existed between leg pairs and between the forehead and nose ridge. A GWAS for the TS confirmed the known QTL at the KIT locus on ECA3, whilst a GWAS for the HS and LS identified novel and distinct QTL indicative of differential trait architecture.

Evaluation of genetic variants proposed as myopathy risk factors in relation to sport performance in riding horses

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Equine myopathies vary widely in frequency, age at onset, types and contexts of clinical manifestation. Genetics play an important role among the causal factors, but diagnostic challenges and involvement of several, possibly interacting mutations complicate scientific evaluations. This study aimed to increase knowledge about six genetic variants proposed as risk factors for muscle integrity myopathy (MIM). Genotype data (MIM test results) were available for 557 Warmblood riding horses (including 219 Oldenburg broodmares) with estimated breeding values (EBV) for sport performance in dressage (D) and show jumping (J) from the national genetic evaluation for riding horses in Germany. At least one mutated allele in > 10 % of tested horses was seen for 4 of the 6 variants (P2, P4, P8, Px). Further statistical analyses considered sport performance through 14 individual EBV and indices (8 for D, 6 for J). General linear models included year of birth, discipline focus in breeding (studbook) and binary variant status (absence vs. presence of at least one mutated allele) as fixed effects. Analyses of variance were performed variant by variant and revealed significant discipline differences (P2, P4, P8, Px; all EBV / indices) as well as some indications of favorable performance potential regarding young horse classes in carriers (P2, P4, P8; up to 6 EBV / indices). The most frequent Px mutation appeared unrelated with genetic performance potential. Further research is needed to verify these findings which may explain observed distributions of mutations referred to as MIM variants.

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Theatre 5

Comparison between SNP array and imputed data to estimate population structure in horse breeds

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The use of BeadChip arrays represents a common tool for studying the genomic structure and diversity of livestock breeds. In contrast, the use of WGS data is expected to improve analysis power. Imputation technique has become standard practice for increasing genome coverage in association studies. This work aimed to extend this approach to biodiversity analysis and compare results before and after imputation. SNP data (40K) from 281 horses (12 breeds) were imputed to sequence level based on a reference panel of 327 sequenced individuals, resulting in ~7M markers after filtering. Both datasets were used to study genetic variability, population structure and runs of homozygosity (ROH). The results showed similar trends, particularly for relationship analyses. Multidimensional scaling and admixture analyses highlighted how the genetic closeness between breeds was increased by the imputation process for those breeds with a low number of sequenced individuals (e.g. Purosangue Orientale Siciliano with Arabian). ROH analysis of the SNP dataset revealed highly shared regions within breeds, and the imputed data gave overlapping results with higher resolution regarding SNP density. In particular, 33 ROH islands on 15 chromosomes were found for the SNP dataset, 25 (on 12 chromosomes) of which perfectly overlapped with the homozygosity patterns detected in the imputed dataset, significantly increasing the total number of markers involved from 876 to 153,198. Five breeds shared the same ROH island on chromosome 11. Further investigation is needed to test the use of the imputation technique in biodiversity analysis. The inferred genomic information can enhance the knowledge of genetic relationships in livestock breeds, paving the way for more robust genomic studies and breeding strategies.

Comparison of genomic signatures of selection in Icelandic horses and Exmoor ponies

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The Icelandic horse and Exmoor pony, both old native breeds, share a history of adaptation to harsh conditions and past population bottlenecks. However, modern breeding practices have distinctively shaped each breed. The Icelandic horse has been selectively bred for performance, while Exmoor pony breeding emphasises maintaining breed standards, particularly regarding exterior features like coat colour and conformation. This study aimed to identify genomic signatures of selection by analysing runs of homozygosity (ROH) islands in both breeds, highlighting their adaptive similarities and unique characteristics. High-density genotype data from 380 Icelandic horses and 274 Exmoor ponies was analysed, revealing shared and private ROH islands in both breeds. ROH islands linked to performance traits were identified in Icelandic horses, characterised by genes such as *DMRT3*, *DOCK8*, *EDNRB*, and *NEURL1*. In contrast, ROH islands unique to Exmoor ponies were associated with coat colours (*ASIP*, *TBX3*, *OCA2*), hypertension (*NELFCD*, *TUBB1*, *GNAS*), immune systems (*LYG1*, *LYG2*), and fertility (*TEX14*, *SPO11*, *ADAM20*). As hypothesized, shared ROH islands between both breeds were related to metabolic processes (*FOXO1*), body size, and the immune system (*CYRIB*). In conclusion, this study illuminates adaptive similarities and the distinct genomic signatures of selection in Icelandic horses and Exmoor ponies, underscoring the impact of divergent breeding objectives on their genetic landscapes.

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Theatre 7

Genomic Dive into Nordic Horse Breeds

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Several Nordic breeds have undergone population bottlenecks, and some are endangered. Limited documentation on the genomic relatedness of these breeds is potentially concealing unique adaptations or detrimental alleles. The aim of this study was to conduct genetic characterization of these breeds to make informed breeding decisions, enhance fertility, and preserve cultural history. Whole-genome sequencing (Illumina NovaSeq 6000) was conducted to an average depth of 18.6 for 190 horses of selected Nordic breeds: Ardenner, Dole, Faroese, Cold-blooded Trotter, Gotland, Norwegian Fjord, Nordland/Lyngen, and North-Swedish horse. The analyses included Fixation Index (*Fst*), Principal Component Analysis (PCA), private alleles, runs of homozygosity (ROH), and heterozygosity. *Fst* values revealed pronounced differentiation, with Faroese horses showing the highest differentiation at 0.25, while Dole and North Swedish Horses exhibited the lowest at 0.04. PCA1 accounted for 20% of the variance, and PCA2 16%, where Dole and Coldblooded Trotter clusters overlapped with North Swedish Horse. Private alleles exhibited hotspots across the genome, with an interesting hotspot at chromosome 20. Inbreeding based on ROH indicated considerable inbreeding, with Faroese horses displaying the highest levels at 0.33. The Dole horse had the lowest observed heterozygosity value of 0.11 and the Swedish Ardennes had the highest of 0.17. Examination of shared haplotypes, effective population size, genetic load, and selection signatures is pending, including comparative analysis aligning the data to Finnhorse assembly.

Genomic characterization of three indigenous Slovenian horse breeds

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The Slovenian Posavje horse (POS), Lipizzan horse (LIP), and Slovenian cold-blood horse (SCH) represent three indigenous breeds with distinct phenotypic characteristics. While the LIP breed is renowned for its elegance, intelligence, and classical dressage abilities and has been extensively researched, the SCH and POS breeds are less studied, despite being the most populous breeds in Slovenia. Recognized for their robustness, adaptability, and calm demeanor, these horses are primarily bred for meat production and also used in agricultural endeavors. Established in 1993 and 2005 respectively, the studbooks for the POS and SCH reflect efforts to conserve these breeds. As part of a conservation program, we genotyped 225 representatives of the POS, 102 representatives of the LIP horse, and 11 representatives of the SCH breed using the Neogen Equine GeneSeek® Genomic Profiler™. Principal component analysis revealed distinct breed-specific clusters, with notable variation within the LIP breed. However, proximity between the POS and SCH clusters indicated a shared genetic heritage between these two breeds. Admixture analysis provided additional evidence of this genetic diversity, suggesting ten inferred ancestral populations while identifying seven genetically different subgroups within the POS breed, and considerable admixture within the LIP breed. Mean ancestry proportions were up to 0.20 for POS (K=8) and up to 0.68 for LIP breed (K=7). These findings suggest a complex genetic structure among and within the studied horse breeds, with evidence of both breed-specific characteristics and shared genetic ancestry, underscoring the importance of comprehensive genetic studies for informing conservation efforts and preserving the unique genetic heritage of these breeds.

Session 48

Theatre 9

Predicting show jumping performance in Belgian Warmbloods using combinations of early life jumping traits

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Show jumping performance is a complex, lowly heritable trait that is only measurable later in life. Hence, studbooks developed early life jumping traits (ELJ) to record jumping capacity at an earlier life stage, either freely (FJ) or under the saddle (JS). Here, we searched for combinations of ten ELJ traits that accurately predict genetic merit for show jumping performance in Belgian Warmblood horses. A total of 2 326 and 2 273 horses scored during FJ and JS contests, respectively, 412 297 horses in the pedigree and 833 497 competition records were used in this analysis. First, we summed all ELJ scores recorded during FJ or JS contests to obtain new phenotypes: ELJIndex(FJ,Sum) and ELJIndex(JS,Sum). Second, we linearly regressed ELJ traits onto estimated breeding values for show jumping performance using ordinary least squares regression (OLS). Using this method, we obtained a combination of ELJ traits and used the regression coefficients to construct ELJIndex(FJ,OLS) and ELJIndex(JS,OLS). Hereafter, we estimated heritabilities of the ELJ indices and computed the genetic correlations with show jumping performance. Estimated heritabilities of ELJ indices ranged from 0.14±0.06 to 0.25±0.04 and were moderately to highly correlated with show jumping performance ($r_g=0.52\pm0.13-0.75\pm0.09$). ELJIndex(FJ,OLS) and ELJIndex(JS,OLS) were better at predicting genetic merit for show jumping performance than ELJIndex(FJ,Sum) and ELJIndex(JS,Sum) (efficiency of indirect selection: 0.91>0.67 and 0.81>0.58). Based on our results, we suggest the implementation of ELJ indices calculated with OLS regression in Belgian Warmblood horses' breeding program as it paves the way for accelerating genetic progress of the population.

Heritability of the ability to race barefoot in Swedish trotters

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In Swedish trotting races, the horses' shoeing condition is routinely recorded. Racing barefoot increases the speed of the horse, but not all horses have hooves that can cope with racing unshod and the hind hooves tend to be the limiting factor. The aim of this study was to estimate heritability for racing with barefoot hind hooves or not ("barefoot status") using repeated observations in Swedish Standardbred trotters (SB) and Swedish-Norwegian Coldblooded trotters (CB). Data from SB races were included from the years 2005-2022 and CB races from the years 2005-2021. The horses were three to ten years old and born 2002-2018 (SB) and 2002-2017 (CB). Because not all trainers race with their horses barefoot, only observations from trainers who had raced barefoot at least once were kept. SB trotters had to have at least ten observations and CB five in the studied period to be included. Barefoot status, a binary trait, included 24,047 SB with 751,723 observations, and 2,986 CB with 74,036 observations. Variance components and heritability was estimated with a univariate threshold model using Gibbs sampling in BLUPF90 suites of programmes. The heritability of barefoot status in SB was estimated to 0.08 ± 0.01 and in CB to 0.09 ± 0.03 with a repeatability of 0.41 in SB and 0.44 in CB. Given that estimated breeding values would be based on several repeated observations, these preliminary results show a potential of including barefoot status in the genetic evaluation of SB and CB as an indirect measurement of hoof quality.

Genetic parameters of summarizing performance traits for evaluating selection approaches for dressage and show jumping sport performance in riding horses

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Successful breeding of sport horses requires breeding programs that meet the challenge of late availability of information on target traits and support early identification and selection of breeding animals. Routines for data collection and analyses of indicator traits are supposed to indicate those mares and stallions with highest probabilities of producing offspring that (1) start a career in sport and (2) perform successfully in competitions up to advanced level. The aim of this study was to determine genetic parameters for summarizing performance traits reflecting these target measures, so allowing to validate criteria currently used to support selection decisions and mating planning. Data of 24,827 mares born 2000-2015 were considered including all active broodmares of the Oldenburg studbooks (OL, OS) plus dams of OL / OS registered foals. Age restrictions ensured coverage with key figures from dressage (D) and show jumping (J) competitions, i.e. total numbers of progeny and their highest levels achieved. Summarizing performance traits were derived per discipline: proportion of progeny with data on highest level achieved in sport (pPS_D, pPS_J), maximal highest level achieved by the progeny (PHEK_D, PHEK_J). Genetic parameters were estimated uni- and multivariately in linear animal models, with results from routine linear descriptions of foals and adult horses providing the data on indicator traits. Linear gait traits for walk, trot and canter (N=12) were analyzed for D, and linear traits for canter and jumping (N=12) were analyzed for J. Heritability estimates for the summarizing performance traits were similar for D and J, ranging from 0.044 to 0.068 ($SE \leq 0.02$). Moderately positive genetic correlations of 0.4-0.6 ($SE < 0.2$) with the directly sport related traits were found for both D (7 gait traits) and J (8 jumping traits). Results indicate the high value of genetic evaluation for linear traits as selection tool in breeding programs for sport horses.

Conformational limbs defects of Pura Raza Menorquina horses: prevalence, heritability and relationship to functionality

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Conformational limb defects are the most prevalent issues in horses, significantly impacting equine performance and welfare. This study examines the prevalence and genetic parameters of 14 limb conformation defects (Open hock, Closed hock, Convergent, Divergent, Camped under, Camper out, Pigeon-toed forelimb, Splay-footed forelimb, Base narrow, Base wide, Coon foot, Broken and upright foot, Sloping foot, Club foot) in Pura Raza Menorquina horses using data from 1,120 records (509 animals) collected between 2015 and 2023. Defects were evaluated using a 3-class scale (0 no defect, 1 slight defect and 2 serious defects), and a Bayesian approach via Gibbs sampling was employed to estimate genetic parameters including all the significant effects in the model: gender (two levels), birth period (three levels), stud selection criteria (four levels), evaluation age (two levels) and appraiser (three levels). Splay-footed forelimb, closed hocks and camped under are the most prevalent defects (67.20%, 62.53%, 55.42%, respectively). Horses with any of the analyzed defects have been observed to obtain significantly lower scores for both walk and trot, with differences of approximately 1/10 point between the defect-free population and the splay-footed forelimb defect. Genetic parameter estimation revealed medium-low heritability values, ranging between 0.16 (s.d.:0.03) open hock and 0.30 (s.d.:0.05) for base narrow. Significant genetic correlations between defects highlight complex relationships requiring careful consideration to include them in the official breeding program. The divergent defect in hind limbs showed high genetic correlations with the forelimb defects camped under (-0.69; s.d.:0.32) and camped out (0.70; s.d.:0.27).

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Theatre 13

From fertility to longevity and climate adaptability: genetics of fitness traits in horses

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The ability to live long and produce foals regularly is crucial for species with low reproductive capacity, such as horses. However, functional longevity is difficult to measure, since it should consider factors enabling individuals to avoid voluntary culling. Fertility, as well, is influenced by many environmental and management factors and few genetic estimates have been done on horses. This study aimed to estimate genetic parameters for fitness traits in Italian Heavy Draught Horse, including: functional longevity (LO), calculated as career length (from the 1st foaling) corrected for morphology and foaling rate; lifetime fertility rate (LFR), expressed on mares' lifetime as number of foals on the opportunities to foal up to the 6th event; the foaling event (FE) or outcome of a mating: 1 when the FE was successful, 0 if not. Each individual had multiple FE records, depending on the number of events. Data was collected over 30 years from approximately 3850 mares and more than 7000 individuals in the pedigree. Single trait models were run to estimate variance components and heritability. A reaction norm model including a temperature-humidity index (THI) covariate was also run on FE based on climate information obtained from the time when mares conceived, allowing to estimate genotype by environment interactions (GxE). Bivariate analyses were run for both fitness traits and morphological traits routinely used. Mares showed a mean LO of 11 years, a mean LFR of 70%, and about 84% of data were successful FE. Heritability was estimated at 0.11±0.02 for FE (liability scale, 0.05±0.01 on observed scale), 0.11±0.03 for LO and 0.24±0.04 for LFR. Heritability of FE increased when GxE were included. Genetic correlations (rg) were moderate among LO and fertility traits (0.16 on average), suggesting the opportunity to include longevity and fertility in a combined fitness index to propose to breeders. High rg (0.74 on average) were found between fertility traits. When GxE were included all rg were reduced and re-ranking in EBVs occurred. Most of the rg with morphological traits were positive. Results suggest the worth of fitness traits and climate information in horse breeding. The Authors thank the ANACAITPR for data.

Structural variants and tandem repeats in horses and their potential impact on withers height

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Structural variants (SVs) are defined as genomic alterations of more than 50 base pairs, which are highly variable in size and type. Just as tandem repeats (TRs), they substantially contribute to the genomic variation between individuals. Therefore, the aim of our study was to investigate SVs and TRs in horses. Variant calling was performed using sequence data from 326 individuals of 49 worldwide breeds, resulting in the discovery of 68,717 autosomal TRs (repeat units ≤ 12 bp) and 33,903 SVs after filtering. To obtain a high-quality call set, the consensus of three callers was used for the SVs. The TRs and SVs were merged with previously called SNPs and INDELs into a multi-type reference panel, which was used to impute 2709 German Warmblood mares from medium-density SNP-arrays to sequence level. Subsequent GWAS for withers height using the imputed data revealed two genome-wide significant association signals on chromosomes 1 and 3, both for the short variants and for the SVs and TRs. These results indicate the possible contribution of SVs and TRs to the variability of withers height in horses. The establishment of our comprehensive multi-variant-type reference panel offers the potential to consider multiple variant types simultaneously in upcoming analyses.

Session 48

Poster 15

Current and Past Development of White Markings in Heavy Warmblood Horses

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White markings (WM) are widespread in domestic horses and are physiologically of high interest because of their early point of origin in embryonic development. Extensive WM or spotting is negatively associated with vitality and health. We aimed to investigate the phenotypical development of WM in the Saxon-Thuringian Heavy Warmblood (STHW) horse for the last 20 years and to use the data for a genetic evaluation to look back into the past. In a first step, a quantitative WM-score was derived from the written WM description for STHW born between 2000 and 2023 (N = 9,519). WM on the head, legs as well as body were summed up into scores for the entire animal (TS), head (HS) and legs (LS). Pedigree information was available with a mean pedigree depth of 5.83 full generations. The heritabilities for TS, HS and LS were 0.72, 0.71 and 0.61 (s.e.<0.03). Finally, EBVs and birth year were available for 19,974 animals, the oldest born in 1900. Based on a linear regression of WM on birth year LSMean, no significant trends could be identified for the TS, HS and LS phenotypes over time. In contrast, significantly decreasing trends were observed in the EBVs of these years for TS, HS and LS. EBVs have the distinct advantage that they can be estimated for animals without phenotypes as long as the animals in question are related to phenotyped animals and thus also enable an evaluation of trends for early years without phenotypes. The evolution of the EBVs seems to indicate that the WM increased significantly in the second half of the last century until the early years of the new millennium. In the first half of the last century, the genetic trend appeared to be near zero.

How to combine microsatellite and SNP for parentage verification in horses?

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Microsatellite markers (MS) have been widely used for parentage verification in most livestock species. In the genomic era, the spread of genotype information as Single Nucleotide Polymorphism (SNP) has raised the question of whether to use SNPs also for parentage testing. Despite the clear advantages of SNP panels, the transition from MS to SNP markers for parentage verification is still very slow and, so far, only routinely applied in cattle. A major drawback is the need for the same genotyping technology for parents and offspring. To overcome this issue, we aim to assess the feasibility of a MS imputation pipeline from SNPs in an Italian horse breed. Over 200 Bardigiano horses were genotyped for seventeen MS and with the SNP chip GGP Equine70k®. Prior to imputation, a quality control is performed, and SNPs located within a window of 2Mb from each MS are selected. The core of the developed pipeline is made up of three steps: a) storing MS and SNP data in a Variant Call Format file, b) masking MS information in a random sample of individuals (10%), c) imputing masked MS based on non-missing individuals (90%). The accuracy of the MS imputation is assessed on the genotype concordance: 0 if none of the imputed alleles matched the true allele, 0.5 if only one of them matched, and 1 if both alleles matched the true alleles. Analyses are ongoing to evaluate if the proposed imputation approach can reach high accuracies in horses. Preliminary results on a subset of 50 horses showed promising results with overall concordance rate above 90% for most of the microsatellites. Michela Ablondi is co-financed by the European Union – PON Research and Innovation 2014-2020 art. 24, par. 3, A) Law 30/12/2010, n. 240 and of the D.M. 10/08/2021 n. 1062.

Session 48

Poster 17

Genomic inbreeding based on runs of homozygosity in the regional German horse breed Schleswig coldblood

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The Schleswig coldblood horse originating from Northern Germany / Southern Denmark was formerly used in agriculture and forestry. With mechanization of these sectors, the number of horses decreased substantially, and the breed was threatened with extinction in the mid-1970s. With around 180 breeding animals, the Schleswig coldblood is today listed as endangered livestock breed (category phenotypic conservation population) by the German Federal Office for Food and Agriculture. The breed has a closed studbook and allows only three other coldblood breeds to maintain genetic diversity: Jutlander, Boulonnais and Southern German coldblood. The aim of this study was to characterize population structure of the Schleswig coldblood in Germany using genome-wide marker data of a large sample from all living horses. Genotypes of 201 horses with 85,401 SNPs were available and used to identify runs of homozygosity (ROH) with PLINK software and its function `-homozyg`. After quality control 57,286 SNPs were retained for analyses with length of DNA stretches considered as ROH set to 2Mb, minimum number of SNPs within one ROH set to 25 and allowing no heterozygotes. For each horse, genomic inbreeding based on ROH was calculated as: $F_{ROH} = \text{total length of all ROH} / \text{autosomal genome length covered by SNPs}$. Mean F_{ROH} was 10.3% (range 0.1-19.9%) in all horses, with no clear trend in inbreeding development over subsequent birth cohorts. However, almost all horses (N=189; 94%) had at least one very long ROH (10Mb), and mean F_{ROH} based on this ROH-length was 3.0%, indicating relevance of recent inbreeding. Systematic use of genomic data for genetic diversity monitoring in the Schleswig coldblood and considering of genomic inbreeding in planning of matings can improve management and support preservation of this breed.

Single nucleotide polymorphisms in four indigenous horse breeds from Poland

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Polymorphism and genetic variation in breeds or populations must be monitored to prevent a reduction in biodiversity and genetic variability. Most often, monitoring is carried out using genetic markers – the same ones used for individual identification and parentage verification. Currently, global trends in research on parentage testing focus on preparing SNP-based methods to replace STRs. The effectiveness of SNP sets for horse parentage testing needs to be investigated. Native Polish horse breeds: the Hucul, Polish Konik, Malopolski and Polish Coldblood (with Sokolski and Sztumski types) are included in genetic resources conservation programs and require monitoring. The aim of the research was to determine the genetic polymorphism, test the panel of 53 SNPs (JPN System), design the own set of SNP markers and determine their usefulness relative to STR markers for selected horse breeds. Both SNP sets, analysed with OpenArray plates on QuantStudio 12K Flex, showed polymorphism in the studied group. Based on the MAF results, 83% of the SNP tested can be used to verify parentage. 100 markers, with MAF above 5%, meet the criteria necessary to create a stable SNP set. The genetic population structure was assessed with Bayesian algorithm to determine if they match the designated breeds. Ten independent simulations for each K value (2-10) assumed a division into four and six horse breeds (encompassing regional types of cold-blood horses). The optimal value was estimated as K=4 in both simulations, confirming the absence of clusters corresponding to regional types and their lack of distinctiveness at the current breeding stage. The SNP sets may be useful also for genetic diversity study.

Session 48

Poster 19

The genomic homozygous landscape reveals selection sweeps in Argentinean Polo ponies

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The Caballo Polo Argentino is a landmark breed recognized worldwide as the most suitable for the practice of Polo. Despite its novelty (the breed was created 40 years ago), an intense selection process relied on the use of embryo transfer and cloning was performed. Here, we analyzed the genomic landscape of 385 polo ponies from 61 different breeders using runs of homozygosity (ROH)-based approach. Horses were genotyped using the Equine-GGPV5 (Neogen, UK). The bioinformatics pipeline included an initial variant call and genotype curation using Genome Studio and Plink. ROH call was performed by a sliding-windows approach in DetectRUNS using the following parameters WindSize:30; minSNP:10; thr:0,05; maxGap:10⁶; mLen:1000000bp, maxOpp:0 and maxMiss:1. ROH analysis and genomic annotation of ROH islands (ROHi) was performed using Bioconductor in R. We detected 42,727 ROH (111 per individual). 80% of the ROH were short (1-5Mb), but 70% of the individuals showed ROH \geq 16.6Mb (associated with very recent inbreeding). On average, the genomic inbreeding coefficient based on ROH (FROH) was 14.3% showing a low correlation with pedigree-based inbreeding coefficient (~0.3, probably associated with weak pedigree records). It also shows a highly variable pattern at the chromosome level. 27% of the FROH was associated with recent inbreeding (probably by the heavy use of large-scale reproductive biotechnology programs). Four ROHi (located in 60% of the samples) were detected in ECA1, ECA7, and ECA17, including 25 genes. Some of them were previously associated with reproductive and metabolic functions in mammals. To our knowledge, this is the first large-scale genomic characterization of Polo Argentino.

Genetic relationship between Neck and Conformational Limb defects in Pura Raza Española Horses for Improved Breeding and Performance Management

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Defects affecting the horse's neck and limb conformation are of paramount importance, since they can negatively impact performance, and horse's suitability for diverse equestrian activities. In this study, the prevalence and genetic correlation between 12 conformational limb defects of the knee, hock, and hoof and 2 neck defects (crest neck -CN-; and ewe neck -EN-) were analyzed in the Pura Raza Española (PRE) horses. Each neck defect was evaluated with the conformational limb defects utilizing a three-class scale (1-no defect, 2-slight defect, 3-serious defect). Significant differences were found in the prevalences of limb defects between the group of animals with or without neck defects. Of a total of 56,644 PRE horses, the most prevalent limb defects in horses affected with CN (20.22%) were splay-footed rear (80.15%) and convergent hock (62.57%), and in horses affected with EN (25.51%), they were splay-footed rear (72.45%) and convergent hock (66.07%). Conformation traits were analyzed with multivariate animal model within a Bayesian framework with BLUPF90 software, incorporating age as covariate, and gender, genetic origin, management of breeder's stud farm and inbreeding as fixed effects. Five generations were used as a relationship matrix (106,971 horses). The genetic correlations between limb defects and CN defect ranged from -0.40(0.03) with convergent hock to 0.30(0.03) with divergent hock. For EN defect, the genetic correlations ranged from -0.23(0.03) for convergent hock to 0.23(0.02) for divergent hock. This study concludes that there is a low-moderate relationship between limb defects and neck defects, emphasizing the need for meticulous planning to improve the studied defects in the PRE breed.

Session 48

Poster 21

Population structure of the Pura Raza Española horse Europe vs America

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The equine industry relies on efficient reproductive and breeding systems for high-quality offspring. Management practices vary by geographical location, but limited studies address these differences. The Pura Raza Española Horse (PRE), found in over 67 countries, is an ideal candidate to do that. This study compares breeding systems and population management for PRE since 1959 across two continents, but due to Spain's substantial equine population, it was analyzed separately. In one hand, reproductive information from 78,601 mares was analyzed (64,380 from Spain, 4,390 from the rest of Europe and 9,831 from America). The reproductive parameters were age at first foaling (AFF, months), age at last foaling (ALF, months), and total reproductive efficiency (RE; the number of total foalings relative to the optimal number of foalings the mare could have during her entire life). On the other hand, population parameters such as inbreeding (F) and average relatedness (AR) were estimated for the total PRE population. In Spain and Europe mares have the AFF one or two months later, respectively, than in America (64 months), but at the same time the ALF is also ten and seven months lower, respectively, than in America (134 months). Nevertheless, RE in America and rest of Europe is similar (38%) and lower than in Spain (40%). From a population point of view, Spain and America have a similar F value (7%) which is higher than in the rest of Europe (6%) while the AR is similar for all locations (11%). Based on these preliminary results, there are differences in breeding systems depending on the country in which the horses are bred, with implications for reproductive efficiency. This information may be of interest to PRE breeders who buy and sell horses around the world.

A protocol to detect early selection criteria for jumping longevity: first results on blood parameters

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In order to find early selection criteria to improve the longevity of show jumping horses in competition, a specific protocol was constructed. Before entering competition, young horses were measured for many traits. These horses were offspring of two groups of sires selected as having the highest and lowest estimated breeding values for functional longevity in jumping competition, as calculated from progeny. Functional longevity was defined as the time spent in competition corrected for the level of performance. The dataset included 952 horses (mainly French Saddlebred) and 77 blood parameters. Heritability was estimated using a mixed model including the effect of age, sex, place and date of collection, weight and animal random additive value with 10,280 horses in pedigree. Heritability of blood parameters was generally moderate to high: 21 values were higher than 0.5 and 39 were between 0.2 and 0.5. The most heritable traits were hematology and enzyme traits: mean corpuscular volume (0.90, se 0.11) and mean corpuscular hemoglobin concentration (0.92, se 0.11) but also traits as liver isozyme (0.72, se 0.13) or total alkaline phosphatase (0.68, se 0.12), small lymphocytes (0.67, se 0.13), superoxide dismutase (0.60, se 0.11). Logistic regression to predict the group of sires favorable to longevity revealed significant effect of lower mean corpuscular hemoglobin concentration (pvalue < 0.001), lower quantity of leukocytes (pvalue < 0.01), lower quantity of liver isozyme of alkaline phosphatase (pvalue < 0.05), higher relative proportion of a2-globulin (pvalue < 0.001) and a1-globulin (pvalue < 0.05), lower relative proportion of monocytes (pvalue < 0.05). Blood parameters measured at rest at young age might be predictive of genetic value for functional longevity in jumping.

Session 48

Poster 23

The possibilities of using genomic information in the selection of mares for milk production

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Mare milk's nutritional advantages are overshadowed by cultural views of horses, which are mainly raised for sports and leisure activities, leading to scant genomic research on its production traits compared to extensive cattle studies. Bridging this gap calls for overcoming cultural hesitations and tapping into markets receptive to horse milk's benefits, underscoring the urgency for more genomic investigations. Lactation efficiency, often dependent on offspring presence, is shaped by genetic factors. Genotype knowledge could aid farmers in selecting mares with superior milk production. Using the commercial GGP Equine chip, we prioritized SNP markers across key gene groups influencing milk production, based on variant consequences, GERP scores, and regulatory significance. Key findings include potentially impactful SNPs for milk production in genes like PRLR, BTN1A1, GH, and GHR. While the SNP chip contains fewer likely causal markers for maternal behavior genes, it includes prospective markers for stress response genes such as CRH, NR3C1, and FKBP5. However, we noted the absence of certain key genes from the chip, notably CSN3, OXTR, and especially DGAT1, highlighting gaps in its coverage for comprehensive genetic studies. Our research aims to design a robust database structure, integrate diverse data sets, and develop technical applications to systematically enhance milk production traits, mirroring methodologies used in cattle research. These data and applications will be organized within the Digitization of Livestock Databases (DigŽiv) project, funded by the Recovery and Resilience Plan and the Ministry of Agriculture, Forestry and Food, to support routine selection work for both breeders and professional services. Moreover, the principles of this approach can be extended to other traits and species. Validation through real data and extensive genetic analyses is crucial for confirming significant markers. Addressing the current lack of genomic efforts in horse production traits is essential for establishing a reference population for milk and other production traits.

Mineral metabolism and health status of dairy cattle fed adjusted mineral supplementation

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Seven dairy herds were selected in the South East of France, with an average of 89 dairy cows / herd. All ingredients of the basal diets including forages were collected monthly and analyzed by ICP-AES for Zn, Cu, Mn, S, Mo and Fe content. For 6 months, cows were fed new mineral feeds, formulated by farm, to provide 80 and 70 mg/kg DM for Zn and Mn, respectively, based on the total diet (+11% and -20% of Zn and Mn supplemented on average). Depending on the level of Cu antagonists in the basal diet (S, Mo and Fe), the target total Cu was on average 11.7 mg /kg DM (-50% on average of Cu supplemented). In each farm, blood samples were collected 3 times on 10% of the animals (n=47; T0=start, T1=3 months and T2=6 months after trial start) and analysed for Zn, Cu and Mn concentrations, as well as the activity of SODE and ceruloplasmin. The effects of the new mineral feeds were explored through ANOVA integrating in the statistical model the treatment (T0 vs T1 vs T2) and the farm as fixed effects and the cow nested within the farm as a random effect. Zinc in plasma significantly increased (P=0.000; T2 > T1 > T0; +21%) while Cu in plasma was maintained with the new mineral feeds (P=0.079; 13.65 µmol/L on average). No statistical differences were observed regarding Mn in plasma for T0 and T2, however, T1 was significantly lower (2.92, 2.82 and 1.96 µg/L respectively). Ceruloplasmin activity significantly increased in T2 compared to T0 (P=0.001; 7628 vs 5708 mU/mL). No statistical differences were observed regarding the activity of SODE (P=0.135). These results showed that Zn, Cu and Mn requirements of dairy cattle can be covered with adjusted mineral feeds without inducing any deficiencies or metabolic disorders.

Session 49

Theatre 2

Nutritional epidemiology sheds light on the effects of alfalfa on dairy cow health, reproduction, and productivity

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Alfalfa offers several advantages in livestock farming. For instance, it can enhance the protein autonomy of farms and outperform straw in preventing acidosis because of its high digestible fiber content (Thomsom et al., 2018). Alfalfa is also rich in fatty acids such as alpha-linolenic acid and in other compounds of interest. In addition, alfalfa might have a positive impact on various health and reproductive parameters of dairy cows. However, experimental evidence supporting these effects remains scarce, mainly because of insufficient statistical power of existing studies. The aim of this study is to quantify the effects of supplementing dairy cows with alfalfa on milk production and composition, animal health, and reproduction. We implemented nutritional epidemiology strategies and analyzed a sizable retrospective dataset. Our dataset covers 580 herds and approximately 100,000 dairy cows over a four-year period (2019 – 2022). Within the dataset, more than 18,000 monthly feed reports offer insights into herd diets, precisely the types of feed and dry matter intakes. In addition, around 1.7 million test day reports delve into milk composition per cow, including fat content, protein content, acetone, BHB, somatic cell count, urea, and milk production. These reports also detail the composition of each cow's milk, covering fatty acids, minerals, and other indicators. Finally, reproduction reports provide information on each cow, such as the calving date, number and dates of artificial inseminations, parity and age. A third of feed reports includes alfalfa. Our ongoing study shows promising results and will be completed by summer 2024.

Effect of sampling method, oral-stomach sampling vs. sampling through ruminal cannula, on pH and concentrations volatile fatty acids and mineral of rumen fluid during an acidogenic challenge in dairy cows
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The use of fistulated ruminants has provided tools for reasoning the feed efficiency of diets, limiting effluents or anticipating the variability in the quality of animal products. Their acceptability by society is called into question. Our objective was to determine whether oral-stomach sampling (OSS) can be an acceptable alternative to sampling through ruminal cannula (Ca) to characterize the variability of rumen fluid composition during an acidogenic challenge in dairy cows. During three one-month-periods, six rumen fistulated cows were subjected to an alternation between a standard diet based on supplemented corn silage (periods 1 and 3) and a diet enriched with starch (period 2). Rumen juice was collected through Ca at three locations in the rumen (reticulum, ventral sac or a mix of both) and by OSS, once per week at 8h30, before morning feeding, and once every third week of each period at 13h30, 4.5 hours after morning feeding. Whatever the sampling method or location, ruminal pH was lower in period 2 compared to periods 1 and 3 at 13h30 (6.21 vs. 5.57 in periods 1 and 2). Ruminal pH was higher when obtained by OSS rather than Ca, whatever the sampling location (+0.44 points at 8h30 and +0.56 points at 13h30). Mineral compositions indicated a presumed dilution by saliva of OSS samples. This was also consistent with lower concentrations of volatile fatty acids. Next steps will be to analyse the associated variations on in vitro gas production and microbiota composition.

Session 49

Theatre 4

Effect of a blend of natural isoquinoline alkaloids on production efficiency, health, and cortisol levels in dairy cows during summer

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The aim of the study was to test a blend of isoquinoline alkaloids (IQs) on production, fertility, and health status of high-producing dairy cows during summer. The trial lasted for 13 weeks and involved 116 cows, allocated into two groups: i) Control, standard diet (n=58, parity: 2.02; days in milk: 51.9); ii) Treatment group, standard diet + 12g head/day of the IQs blend (n = 58, parity: 2.07; days in milk: 48.8). Milk yield, dry matter intake (DMI), feed conversion rate (FCR), body condition score (BCS), milk quality, apparent total tract digestibility (aTTD), reproductive and health parameters were evaluated. Blood cortisol levels were analysed on 10 cows per group at the start (d0) and after 60 days (d60). All the data were analysed using the SAS software. The average milk production (+0.85 L/head/d) (P<0.0001) and FCR (1.43 vs 1.38) (P<0.0001) were significantly higher in the Treatment compared to Control group, starting to differ from week 9 and 8 respectively. No effects were detected on DMI, milk quality parameters, BCS and aTTD values. In terms of fertility, the average number of services was reduced (1.93 vs 2.34 service/animal) (P=0.0028) while a higher percentage of cows were pregnant at the end of the trial (72.41 vs 37.93%) (P=0.0001) in the Treatment group. The total incidence of diseases was lower in the Treatment group (13.33 vs 50% of affected animals) (P=0.0010). The cortisol levels were improved in the Treatment group at d60 (0.339 vs 0.586 µg/dL in the Control) (P=0.0032). The administration of a blend of IQs during the summer months have shown positive effects on production efficiency and overall health, enhancing animal resilience.

Bacillus coagulans supplementation in Holstein Friesian female calves' diet

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After birth, twenty animals were divided into two groups: a control group (CTR) fed a basal diet, and a treatment group (T) fed a concentration of 10⁹ CFU/kg of Bacillus coagulans DSM32016 (Technospore®). Trial started at 7 d of life (0 d of trial) and lasted 56 d. Colostrum was sampled at 6-12 hours after birth. Body weight (BW) evaluation, blood, and fecal sampling were performed at 0, 28 and 56 d. Plasma glucose, non-esterified fatty acids (NEFA), beta-hydroxybutyrate (BHB), IgG and serum total antioxidant capacity (T-AOC) were analysed. Data were analyzed with a two-way ANOVA and a Bonferroni test. Fecal samples were analyzed through 16S rRNA-gene sequencing. MetaCyc database was used for functional prediction, followed by a Bonferroni-Dunn evaluation. Colostrum IgG showed levels above 50 g/l. BW was higher in T vs CTR at 56 d (p<0.05). Plasma metabolites were not conditioned by the treatment. Serum T-AOC increased in T vs CTR at 56 d (p<0.05). β-diversity revealed differences between T and CTR at 28 and 56 d (p<0.05). Bifidobacterium, Prevotella 9 and Fusibacterium were more abundant in T vs CTR at 56 d (p<0.05). Starch degradation and L-isoleucine biosynthesis pathways significantly increased over time in T and were correlated with the cited taxa at 56 d (p<0.05). The supplementation of Bacillus coagulans DSM32016 ameliorated growth and serum T-AOC of calves, revealing its capacity to modulate potentially useful microbial niches.

Session 49

Theatre 6

Effects of live yeast Saccharomyces cerevisiae boulardii on gut permeability and milk performance in transition in dairy cows

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Inflammation in dairy cows, during the transition period, is thought to be related to increased gut permeability, and could be a source of impaired performances in early lactation. The aim of the study was to investigate if the supplementation of Saccharomyces cerevisiae boulardii CNCM I-1079 (SCB) could alleviate these challenges. Eighty-three multiparous (n = 52; MP) and primiparous (n = 31; PP) cows were blocked by previous 305-d milk yield, parity, body condition score (BCS), and body weight (BW), and randomly assigned to either prepartum and postpartum control (CON; n = 43) or SCB (n = 40) dietary treatments. SCB had no effect on dry matter intake (DMI) but increased 3.5% fat-corrected milk (FCM) and energy corrected milk (ECM) yield. Milk fat content and yield were greater with SCB. Ruminal indigestible marker Cr-EDTA was dosed for gut permeability assessment. Cr area under the curve (AUC) was greater on day 7 compared than day 70 postpartum, indicating increased permeability during early lactation. SCB tended to reduce Cr AUC, suggesting improved gut integrity. These results suggest SCB may enhance milk performance due to limiting the gut leakage during the transition in dairy cows.

Reducing the omega-6-to-omega-3 ratio in milk replacers modulates feed intake behavior and growth in calves fed ad libitum

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Milk replacers (MR) for calves have a higher omega-6 (n-6) to omega-3 (n-3) ratio than whole milk because of the use of alternative fat sources to milk fat, which can modulate metabolic responses. This study investigated the effect of reducing the n-6:n-3 ratio in MR on growth and feeding behavior. Thirty-six Holstein calves (2.8 ± 1.06 d old) were blocked based on arrival day. Within each block, calves were randomly assigned to a control MR with an n-6:n-3 ratio of 40.6:1 (CON, 65% palm and 35% coconut fats, n=18) or an MR (n-3, n=18) in which linseed (1.2%) and algae oils (1.3%) were incorporated in the same fat blend than CON resulting in an n-6:n-3 ratio of 9.8:1. Both MR were isoenergetic with 29% crude fat, 24% CP, and 34% lactose (DM basis). Calves were group-housed and fed ad libitum MR between wk 1 and 6, gradually weaned between wk 7 and 10, and fed only solid feed between wk 11 and 12. Starter feed, straw, and water were provided ad libitum. Calves were weighed weekly, and feed intake behavior was recorded daily using electronic recognition. Continuous variables were analyzed using a mixed-effects model with repeated measures. Milk replacer intake was greater in calves fed CON ($P < 0.01$), resulting in a trend for enhanced BW throughout the study ($P = 0.06$). Calves fed CON had a higher number of unrewarded visits to the automated milk feeder during weaning ($P = 0.04$), whereas there were more interrupted milk meals for calves fed n-3 throughout the study ($P = 0.01$). Differences in feed intake behavior suggest a direct link between the n-6:n-3 ratio in MR and satiety regulation.

Session 49

Theatre 8

Incorporation of butyric and caproic acids in milk replacers modulates postprandial metabolism in calves

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Using alternative fat sources to milk fat in milk replacers (MR) for calves leads to low levels of butyric and caproic acids. This study investigated the effects of incorporating tributyrin (TB) and tricaproin (TC) into the MR on postprandial metabolism. Forty-five Holstein calves (2.1 ± 0.63 d) were blocked by arrival day. Within each block, calves were randomly assigned to MR treatments (15 calves each) differing in fat composition: (1) milk fat (MF), (2) vegetable fats (CON), and (3) an MR with the same fat blend as CON, to which TB and TC were incorporated (TRI). All MR contained 36% lactose, 27% fat, and 24% CP (DM basis). Calves were housed individually, and daily milk allowances were divided into 2 meals (135g/L), which were gradually increased to 8L/d on d10. Postprandial blood sampling took place on d21 over 7.5h relative to the morning meal, which contained acetaminophen (Ac), to measure abomasal emptying. Continuous variables were analyzed using a mixed-effects model with repeated measures. The area under the curve (AUC) for Ac was lower in calves fed TRI than other treatments ($P = 0.05$). Postprandial insulin (ins) was lower in calves fed TRI than other treatments, whereas glucose (glc) was not different. The AUC_{glc}:AUC_{ins} ratio tended to be greater in calves fed TRI than other treatments ($P = 0.07$). Postprandial triglyceride (TG) were higher in calves fed MF than in those fed TRI at 30, 120, 150, and 360 min post-meal ($P \leq 0.03$), whereas CON did not differ. At 180 and 420 min post-meal, postprandial TG was greater in calves fed MF than other treatments ($P \leq 0.05$). Incorporating TB and TC into MR slowed down abomasal emptying, modulated postprandial metabolism, and enhanced insulin sensitivity.

Evaluating the effects of rumen originated lipopolysaccharide on the pathogenesis of rumen acidosis

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Subacute ruminal acidosis (SARA) is a well-recognized digestive disorder of high yielding dairy cows that has a negative impact on both animal health and herd profitability. It is associated with increased ruminal lipopolysaccharide (LPS). Our previous data showed that LPS from *E. coli* (LPS-E) increased the starch-digesting and lactate-producing ruminal bacteria. However, it is not known if LPS-E would be representative of LPS from ruminal bacteria (LPS-R) to evaluate its effect on the pathogenesis of SARA. Therefore, the objectives of this study are to 1) collect LPS released from the rumen bacteria by inducing the SARA nutritional model in batch culture; 2) compare the effects of LPS-R with the LPS-E on bacterial community composition (BCC), and function by using 16S rRNA sequencing and RNA-seq. The SARA was induced by cultivating rumen mixed bacteria with TMR containing 40% starch in 1/12 Menke & Steingass's buffer, which was able to maintain the pH < 5.6 for more than 180 min. LPS-enriched culture media (LPS-R) were collected 20 hours post-incubation using centrifugation and 0.22 µm filtration, aligning with peak LPS concentration (22,172 EU/ml) during a 24 h incubation, which was ~ 7 times higher than non-SARA group (3,244 EU/ml). LPS-E and LPS-free media were adjusted to match the LPS, VFA, and lactic acid concentrations of LPS-R. All media were prepared with a TMR substrate, essential minerals, vitamins, and adjusted to a final pH of 6.8. Each medium was dispensed into 8 ml aliquots within Hungate tubes. The rumen mixed bacteria 20% of the cultivated volume inoculated in these media. Bacterial samples were taken at 6 h and 24 h after inoculation. Ongoing 16S and RNA-seq analyses seek to elucidate their BCC and specific functions across treatments.

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Theatre 10

In vitro effects of mycotoxin Enniatin B on bovine PMNs immune function

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Enniatin B is an emerging mycotoxin that can have a potential toxicity on the innate immune response both in animals and humans. This study investigated for the first time the effect of increasing ENN B concentrations (0.625, 1.25, 2.5, 5, and 10 µM) on bovine polymorphonuclear leukocytes (PMNs) in vitro. PMNs viability, chemotactic function, phagocytic ability, and extracellular Reactive Oxygen Species (ROS) production were determined with or without phorbol myristate acetate (PMA) as a pro-inflammatory challenge. Dimethyl sulfoxide (DMSO), was used as a vehicle to solubilize ENN B and included as a negative control. Data were analyzed by GraphPad Prism and normality was assessed by Shapiro-Wilk test. Repeated measures of 1-way ANOVA for matched or paired data and Tukey's multiple comparison test were then applied on the considered parameters. PMNs viability and chemotactic activity were not affected by ENN B at all tested concentrations ($p=0.952$; $p=0.218$, respectively), while *E. coli* and *S. aureus* phagocytosis ability were reduced by the highest concentrations (ENN B 5 and 10 µM) compared to DMSO ($p\leq 0.001$; $p=0.001$, respectively). Extracellular ROS production was increased by ENN B under normal and pro-inflammatory conditions ($p=0.014$; $p<0.001$, respectively). In conclusion, ENN B did not exert cytotoxic effects on bovine PMNs, while it reduced the phagocytic ability and increased the production of extracellular ROS highlighting its potential role on bovine innate immune response.

Phenolic Compounds in Animal Nutrition and Health: Chronic Kidney Disease

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Chronic Kidney Disease (CKD) poses a significant global health challenge, necessitating innovative and accessible treatment modalities. This review investigates the therapeutic potential of phenolic compounds (PC) in treating and managing CKD in animals and its associated symptoms. This review delves into the current studies on phenolic acids, flavonoids, tannins, and other noteworthy compounds, exploring their multifaceted biological activities. Our findings highlight the potential of these compounds in addressing CKD-related issues, such as antioxidant, anti-inflammatory, and potential anticancer effects, as well as their role in managing associated conditions like diabetes and hypertension disorders. In addition, our research also comprehensively analysed the nutritional benefits, bioavailability, toxicity, and safety of PC while discussing the implications of our findings for animal studies. We emphasise the urgent need for further research to validate the therapeutic potential of PC for animals with CKD by using it as a supplement to animal diet while acknowledging the challenges and limitations of current research. This sets the stage for future research on PC in CKD. In conclusion, our work presents a compelling call to action for the scientific community to explore the utility of PC in addressing CKD-related challenges, particularly in the animal and nutraceutical industries. Acknowledgements: This work is supported by National Funds by FCT – Portuguese Foundation for Science and Technology, under the project UIDB/04033/2020 (<https://doi.org/10.54499/UIDB/04033/2020>).

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Protective effect of chlorogenic acid on liver injury in heat-stressed meat rabbits

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This study investigated the protective effects of chlorogenic acid (CGA) on production performance and liver function of rabbits under heat stress (HS) condition. A total of 120 healthy New Zealand weaned rabbits with similar initial body weight, were randomly divided into 3 treatments with 20 replicates per treatment and 2 weaned rabbits per replicate: control (CON) group (rabbits were housed at 25±1°C and fed a basal diet), HS group (rabbits were housed at 35±1°C and fed a basal diet), and HS + CGA group (rabbits were housed at 35±1°C and fed a basal diet supplemented with 800 mg/kg CGA). The trial lasted for 28 days. The results showed that HS challenge decreased ($p<0.05$) growth performance, induced oxidative stress and hepatic apoptosis, and caused liver damage in rabbits. However, dietary CGA supplementation increased ($p<0.05$) body weight gain and feed efficiency, and enhanced ($p<0.05$) antioxidative capacity in serum and liver in HS-challenged rabbits; attenuated ($p<0.05$) HS-induced increases in urea nitrogen, alanine aminotransferase, aspartate aminotransferase, caspase-8, and caspase-3 as well as decrease albumin to the levels observed in CON group. Moreover, supplementation with CGA upregulated ($p<0.05$) Nrf2/HO-1 pathway-related genes (Nrf2, HO-1 and SOD1) expressions in HS-challenged rabbits. Our findings demonstrated that dietary CGA supplementation could alleviate HS-induced decline in growth performance, and protect against HS-induced liver damage partially through enhancing antioxidant capacity via acting Nrf2/HO-1 pathway and inhibiting hepatic apoptosis in rabbits.

In vitro nutritional interventions to enhance rumen fermentation during the post-weaning process

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Low growth rates and digestive disorders are common problems during the post-weaning process in artificially reared lambs given their immature rumen. This study aimed to assess the effects of feed additives on in vitro rumen fermentation profile and gas production (GP) using rumen inoculum from young lambs. Four artificially reared lambs were used as donors, and inoculum was incubated for 24h in batch cultures containing a high concentrate diet (92%). The six commercial feed additives were: fatty acids (FA) treatments including medium-chain (lauric acid) and long-chain FA (halibut liver oil), oleins (saturated vs polyunsaturated), probiotics (live yeast) and prebiotics (pectin). Additives were included at four doses (0, 0.1, 0.3, and 0.6 g/L). Result showed no significant differences for incubation pH and concentration of total volatile fatty acids (VFA). Lauric acid and live yeast had no effects on the rumen fermentation pattern. The addition of oleins decreased the GP during the late fermentation, suggesting certain anti-microbial properties. Oleins rich in saturated FA promoted a shift from propionate to butyrate production, whereas oleins rich in polyunsaturated FA decreased methane production during the late fermentation. Halibut liver oil accelerated the GP. Pectin supplementation resulted on the most promising strategy as it linearly increased GP though and decreased rumen ammonia concentration. These results indicated that the modulation of the rumen fermentation of artificially reared lambs is challenging, possibly as a result of the low microbial development and low rumen pH. However, the supplementation with prebiotics such as pectin might slow down the rumen fermentation process leading to positive effects under these physiological conditions.

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The form of administration and dose of astaxanthin on calves' growth and health

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The aim of study was to determine the form of administration and dose of astaxanthin (AXT) on the development and health of calves during the period of feeding with milk replacer. 32 pHF calves, divided into 4 groups, 8 animals in each. Group I (control) received milk replacer and concentrate mixture, group II received AXT as a supplement to the concentrate mixture (20 mg/kg DM), groups III and IV received AXT in milk replacer in the amount of 0.11 mg/kg bw/day and 0.25 mg/kg bw/day, respectively. AXT dissolved in a small amount of milk replacer was administered to the calves. The experiment was carried out until the 56th day of the calves' life. During the experimental period, the body weights and body weight gains of calves were determined. Blood was collected on the 1st, 7th, 14th, 28th, and 56th day of the calves' life. The obtained data were analyzed using Statistica 13.1 (2021). The daily weight gain of calves in the groups receiving AXT in the milk replacer was higher than in the control group and group II receiving AXT in the concentrate formula. Calves from group III consumed almost half as much ($P \leq 0.01$) AXT compared to groups II and IV (5.25 mg/day). Effect was found in glucose, NEFA, BHBA ($P \leq 0.05$). In the groups receiving AXT in milk replacer, higher values of glucose and the beneficial HDL cholesterol fraction, as well as lower values of lactate dehydrogenase, were found. A higher content of P and Mg was found in the blood serum of calves receiving AXT in dissolved form. The addition of AXT had a positive effect on the level of immunoglobulins ($P \leq 0.01$). Experiment conducted on calves up to 8 weeks of age showed that AXT influenced the body weight of calves, biochemical parameters and blood effects. Astaxanthin dissolved in milk replacer is better absorbed by calves. The optimal AXT doze for calves is half of that recommended for adult ruminants.

Development of suitable exterior indicators of milk-deprived and milk-fed suckling calves dependent on age
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Indicators for the nutritional status and body condition of suckling calves are currently missing but necessary to ensure animal welfare in “cow-bonded calf rearing systems”. To develop an assessment, expert interviews (n=11) with experienced (> 20 years) people from the dairy sector were conducted via the video conferencing service Zoom. Based on the literature search and suggestions from the interviews an evaluation protocol for suckling calves was developed, which was tested at the Bavarian State Research Farms (BaySG) Achselschwang and the experimental farm Veitshof of the Technical University of Munich (TUM). Ninety-eight calves aged from birth to six weeks of life were scored regarding 46 criteria in milk-deprived and milk fed state. The influence of age on the body parameters was assessed using the Kruskal-Wallis-Test and nutritional status using the Wilcoxon-Test. Receiver operating characteristic curves were determined by the Youden index. Most promising was flank (width and circumference), which was enhanced with increasing age and nutritional status. Dewlap (length and width), shoulder width and chest girth display the age effect. With the threshold values found for different ages, it should be possible to predict (50- 96,4 %) the nutritional status of a suckling calf with known age in the first three weeks of life.

Survivability of LACTIFERM® as a direct fed microbials for calves: application to feeding practices
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Under the last couple of decades, direct fed microbials (DFM) have been increasingly used in animal production. In young ruminants, DFM were linked to improvement of body weight and size, feed efficiency, resistance to diseases, establishment of the rumen microbiota as well as development of rumen papillae and function. However, prior to reach the gastrointestinal tract and exert their beneficial effects, DFM need to survive to manutention, transport, and wide diversity of feeding practices. In this study, we investigate the survivability of LACTIFERM® (Novonosis, Denmark), a commercial product for calves, in milk replacer at room temperature, in milk replacer acidified with formic acid as well as its compatibility with halofuginone, a veterinary drug used for cryptosporidiosis in newborn calves. The capacity of *Enterococcus faecium* 669, the active strain from the LACTIFERM® product, to grow in a complex medium mimicking the composition of rumen environment after a meal or in pure rumen fluid was assessed by optical density. LACTIFERM® displayed an excellent survivability and growth ability in milk replacer left at room temperature over 24 hours as well as excellent stability on acidified milk replacer with formic acid. Simulation of halofuginone supplementation after a meal of milk replacer did not impact the multiplication of LACTIFERM® at calf stomach temperature. At last, *E. faecium* 669 demonstrated the ability to grow in rumen containing media. Altogether, this study suggests that LACTIFERM® possess the capacity to deploy its full beneficial properties in calf rumen even while handled with various feeding practices.

Daily fluctuation in voluntary dry matter intake effects on feedlot performance of Nelore cattle

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This study aims to evaluate how day-to-day fluctuation (DF) in voluntary dry matter intake (DMI) influenced feedlot performance of 123 Nelore bulls (15±3 month of initial age and 355 ± 5.90 kg of initial body weight). Animals were fed high concentrate feedlot diets for an average of 90 days. DF was calculated by the difference between DMI of the previous and the current day [%DF-DMI = ((DMIPrevious – DMICurrent) * 100) / DMIPrevious], expressed as percentage. Animals were classified as low (L; n=71 animals) or high (H; n=52 animals) DF-DMI groups: DF of 1.45 and 2.84%; or 0.88 and 2.25 kg/d, respectively (P<0.001). Data were analyzed by SAS MIXED procedure considering as fixed effects DF (L or H) and age at slaughter as covariate and the random effect of diet. Means were compared by F-test with α=0.05. No differences were detected between L and H DF-DMI groups for average daily weight gain (1.11 x 1.12 kg/d, P=0.85) and DMI (9.12 x 9.16 kg/d, P=0.35), respectively. However, animals within H DF-DMI group had lower slaughter body weight (SBW; 458 x 475 kg, P=0.04) and hot carcass weight (HCW; 269 x 281 kg, P=0.03) than the ones within L DF-DMI group. Also, trends towards greater back fat thickness (4.53 x 4.14 mm, P=0.06) and rib eye area (84.1 x 80.8 cm², P=0.06) were detected for L DF-DMI animals when compared to the H DF-DMI ones. The fact that reduced DF-DMI was related to greater SBW and HCW makes this measurement relevant as an index of feedlot performance of Nelore cattle. Reducing DF-DMI should improve cattle health and performance.

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Poster 18

TMR quality preservation supports milk production performance

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The aim of this study was to evaluate at the end of summer, the production performance through the preservation of TMR quality. The field trial was carried out at a high-production dairy farm in northern Italy. The farm consists of 120 lactating cows, divided into multiparous (MP) and fresh cows and a group of primiparous (PP) and high DIM cows. The TMR was provided twice a day and the cows were milked three times a day by the robot. The trial period lasted 60 days. The production results of a control group (days 1-30: PP cows; days 30-60: MP cows) and treatment group adding 2.0kg/MT of TMR of a synergistic blend of organic acids for TMR preservation (days 1-30: MP cows; days 30-60 PP cows) were compared. The data measured were average milk production, ADI weighing the excess TMR at end of day, THI, milk quality, and TMR temperatures. The results showed differences between the treatment and control groups, even though average ambient temperature was 24°C and THI never exceeded 72. The TMR temperature remained higher in the control groups, with peaks on day 19 and 51 with > 10° degrees difference after 8 hours of TMR production. Feed waste increased in day 19 and 51 in the control group, with a total average of 1.6% in the control group and 1.3% in the treated group. There were no significant differences in milk quality and solids, nor in feed efficiency (1.58 control vs 1.61 treated), the greatest effects emerged at the level of average milk production +0.9 kg/h/day of milk with a significantly difference, (p-value = 0.0005) between control and treated group. The use of organic acids blend (Selko® TMR Dry Selko®, Tilburg, The Netherlands), supported the quality preservation of TMR in non-extreme heat conditions in a farm with good management practices regarding hygiene, with feed ratio provided twice a day.

Effect of a blend of essential oils on rumen microbiota for relieving sub-acute ruminal acidosis

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The study aimed to evaluate the effects of a commercial blend of essential oils (EO) on the rumen microbiota under subacute ruminal acidosis (SARA) condition. Four dry rumen-cannulated cows were involved in a cross-over design experiment consisting of two experimental periods with 21-d adaptation fed either Control diet (CON) or CON + 3 g/d EO. At the end of the adaptation period, rumen fluid from each cow was collected and allocated to two substrates for 24-h in vitro incubation: Control diet (CON_TMR, 18% starch) and SARA diet (SARA_TMR, 46% starch). The SARA diet maintained the pH \leq 5.6 for more than 3 h/d. At the end of incubation, the culture media were collected to count rumen protozoa and harvest rumen bacteria for 16S rRNA sequencing. The 16S rRNA gene sequencing data was analysed using DADA2 software, assigned taxonomy using SILVA v132 databases and phylogeny was generated by QIIME2. Supplementation of EO reduced the total number of rumen protozoa ($P < 0.01$). However, no effects were observed on the α and β diversity of rumen bacteria community composition. Supplementation of EO decreased the relative abundance of Methanogens while increased the relative abundance of fibre-utilizing bacteria, such as Ruminococcus ($P < 0.05$). After supplementation of EO, the α and β diversity of rumen bacteria community composition were not different between CON_TMR and SARA_TMR. However, the relative abundance of Streptococcus was numerically decreased when inoculated in EO substrate compared to CON. In conclusion, supplementation of EO could relieve SARA by improving the fibre utilizing bacteria and potentially decreasing Streptococcus in the rumen.

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Poster 20

Bromide: A water quality constituent of concern

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Extensive livestock production systems across South Africa often rely on groundwater sources for livestock. South Africa lies in the subtropical zone of southern Africa that has a range of ecosystems which include semi and desert regions. The subtropical region is prone to drought that may last several seasons, dry seasons and variable meteorological conditions. In addition, extensive livestock production is not a totally free-ranging system, since controlled grazing and herbage usage are fundamental to sustainable range management practices. The consequence is that livestock watering relies to a large extent on groundwater. The official South African Water Quality Guidelines (SAWQG): Agricultural Water Use: Livestock Watering (1996) does not include Bromide (Br-1) as a water quality constituent of concern (WQCC). Several subsequent analyses of groundwater have reported Br-1 occurring in concentrations that could be considered a WQCC. These have been reported in research papers and reports of the S. A. Water Research Commission. Results of Br-1 concentrations having negative effects on the development of highly sensitive chicken embryos raise the question of the extent to which Br-1 could be considered a WQCC. The risk posed is the development of embryos of gestating and egg-producing animals exposed to acute or chronic concentrations of Br-1 via groundwater, could be affected negatively. This paper presents the potential effects of Br-1 on livestock production and the need to include Br-1 as a WQCC in the SAWQG.

Evaluation of tannins supplementation as functional feed additive on neonatal diarrhea in calves

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The aim of this study was to evaluate the in-milk supplementation of chestnut and quebracho tannin extract with or without saponins combination on performance, diarrhea frequency, and protein digestibility in neonatal calves. Trial 1 involved twenty newborn calves divided in two groups: control group was fed whole milk (n=10; CTRL) and tannins group (n=10; TAN) was fed whole milk supplemented with 6 g/day of tannin extract (Silvafeed ByPro) for 30 days. Calves' weight was registered at 0, 14 and 30 days. Fecal samples were obtained at 3, 7, 14 and 30 days for analyzing the protein digestibility. Fecal consistency was evaluated using the fecal score scale (0–3 scale, considering diarrhea > 1). Trial 2 involved 14 calves divided into control group (n=7; CTRL) fed milk replacer and tannin and saponins group (n=7; TANSAP) fed milk replacer supplemented with 5 g/day of tannin and saponin extract (Silvafeed BX) for 28 days. Animals were weighed at 0, 14 and 30 days. Feces were collected at 0, 14 and 28 days for the evaluation of protein digestibility and daily scored to evaluate diarrhea frequency. Blood samples were collected at 0 and 28 days for the evaluation of antioxidant barrier through colorimetric test. Additives supplementation did not influence the growth performance and protein digestibility without altering the palatability of milk. Tannins supplementation significantly decreased diarrhea frequency compared to CTRL groups ($p < 0.05$). TANSAP group showed significantly increased the antioxidant barrier at 28 days compared to CTRL in Trial 2 ($p < 0.05$). In conclusion, the in-milk supplementation of tannins could be considered a valuable strategy to decrease diarrhea occurrence, thus supporting animal health and decreasing antibiotic use in livestock.

Does *Saccharomyces cerevisiae* postbiotic affect hay in vitro digestibility? A comparison between inclusion in the rumen fluid inoculum and dietary supplementation in the donor Chianina bullocks

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Postbiotics are emerging to promote a favourable ecosystem for bacterial growth. In vitro 48 h fermentations were performed in the Ankom–Daisy II incubator to evaluate the effect of a postbiotic based on *Saccharomyces cerevisiae* (Doxal Italia S.p.A.) on rumen microbiota ability to degrade *Medicago sativa* hay. pH was recorded after 0, 24 and 48 h of fermentation. In experiment 1, fermentations were performed with non-treated rumen fluids collected at the slaughterhouse from 6 Chianina bullocks. The two-way ANOVA indicates that the direct inclusion of two postbiotic doses (5 and 10 mL) did not affect dry matter (DM), neutral detergent fibre, and crude protein digestibility. The pH value at 48 h was increased ($P < 0.05$) by the postbiotic inclusion at the higher dose (6.8 vs. 6.6). In experiment 2, the rumen fluid used for the in vitro fermentation was obtained from Chianina bullocks fed either a control diet or the same diet supplemented (2% DM) with the postbiotic. Rumen fluid was collected through a rumen probe at 0, 30 and 60 days of postbiotic dietary administration and incubated as in experiment 1. Statistical analyses showed no differences in digestibility and pH values. Further in vitro and in vivo studies with genomic approaches are warranted to better elucidate the postbiotic effect on the rumen ecosystem.

Assessing the colostrum proteome of Holstein cows, and its effect on calf passive immunity acquisition
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The aims of the study were to explore the proteomic profile of bovine colostrum, to examine how colostrum IgG levels and pre-calving blood globulins may impact this profile, and to evaluate associations between the colostrum proteome and calf acquired passive immunity. Colostrum samples of 18 Holstein cows were classified in Poor, Average and Excellent quality based on colostrum IgG. Based on pre-calving blood globulins, blood samples were classified in Low, Moderate, and High. Calves' passive immunity was estimated at 7 d of age. The colostrum proteome was evaluated with mass spectrometric analysis using LC, which was directly coupled to a high resolution Q Exactive HF Orbitrap mass spectrometer. On average, 285 proteins were detected in colostrum samples, representing primarily the biological process for immune system response. C4b-binding protein alpha chain, CD5 molecule like C4, complement component, and serpin A3-7 were upregulated in colostrum samples with Excellent quality. However, beta and alpha-S1-casein, xanthine dehydrogenase/oxidase, and HGF activator were down regulated. This resulted in enrichment of innate immune response and negative regulation of endopeptidases. Pre-calving blood globulin content correlated with abundance of several colostrum proteins, such as IgG and lactotransferrin. Furthermore, certain colostrum proteins, such as fructose bisphosphate aldolase and peroxiredoxin 1, correlated with calf acquired passive immunity. This study provides new insights into the colostrum proteome, showing influence of IgG level on other proteins. There were correlations between pre-calving blood globulin and colostrum proteins. Correlations between different colostrum proteins and calf passive immunity suggest that assessment of passive immunity transfer should consider the content and interaction among the myriad of colostrum proteins.

Changes in calves' immune response affected by herbs and their blends – *ex vivo* study
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Calves in the first weeks of life, due to the immaturity of their immune system, are most susceptible to diseases. Immunoprophylaxis may replace antigenic stimuli necessary for the faster maturation of the calves' immunity. Therefore, there is growing interest in modulating the immune system with plant-derived products such as phytoimmunostimulants, which include herbs. The study aimed to explain the mechanisms involved in the regulation of the immune system in calves. The research investigated the effect of single herbs extract exhibiting immunomodulating activity (thyme *Thymus vulgaris* L., purple coneflower *Echinacea purpurea* (L.) Moench., sage *Salvia officinalis* L., rosemary *Salvia rosmarinus* L. and cinnamon *Cinnamomum verum* J. Presl) or their mixtures on the stimulation of the calves immune response *ex vivo*. The blood samples from 8 newborn healthy calves were collected and cells were cultured for 4 days. Evaluation of specific immunity involved testing lymphocytes isolated from peripheral blood using a concentration gradient stimulated or not in a complete culture medium in the presence of mitogens. The determinations of markers of differentiation were carried out by flow cytometry, which allowed for accurate analysis of immunophenotyping of calf peripheral blood lymphocytes. The results of the observations showed different activations of lymphocytes depending on the herb used and their concentration. Increased activation of markers and lymphocyte proliferation were observed. In addition, changes were observed in the proportions of different subpopulations of lymphocytes. It suggests that the use of herbal extracts may have the potential to stimulate the immune response. Further research into the use of immunomodulatory herbs and their efficacy *in vivo* may be a promising next step for developing nutritional strategies to support immunity in newborn calves.

Effect of supplementing a *Saccharomyces cerevisiae* fermentation product from dry-off through 90 days of lactation on the metabolic adaptations of dairy cows with previous lactation high somatic cell count
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A group of 14 Holstein cows having high SCC (> 100 K/mL for primiparous and > 200 K/mL for multiparous cows) were enrolled 20 days before dry-off. Cows were then assigned to two homogeneous groups and received diets supplemented with 19 g/d of a *Saccharomyces cerevisiae* fermentation product (SCFP; NutriTek®, Diamond V) or without supplementation (CTR) until 90 days from calving (DFC). Disease incidence, milk yield and composition, plasma metabolic profile and whole blood count were monitored. Data were analyzed with ANOVA and mixed models for repeated measures. Significance was defined as $P < 0.05$. During dry period, compared to CTR, SCFP cows had reduced plasma protein, globulin, haptoglobin, ceruloplasmin and reactive oxygen species to ferric ion reducing antioxidant power ratio, greater plasma concentration of albumin and thiol, heightened albumin-to-globulin ratio, hematocrit and greater concentration of leukocytes and lymphocytes. After calving, SCFP cows had a lower mastitis incidence, lower SCC between 1 and 7 DFC, higher butterfat, greater ECM between 22 and 60 DFC, greater blood hemoglobin and plasma haptoglobin, gamma glutamyl transferase, glutamate oxaloacetate transaminase and Zn, and reduced plasma Mg compared to CTR. The ameliorated immune system function lowered the SCC and prevented the onset of new intramammary infections at the onset of the new lactation in SCFP cows, improving their productive performances.

Session 49

Poster 26

Effects of supplementation with vitamin E or plant extracts on immune and mammary epithelium integrity after a LPS challenge in lactating dairy cows

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Early lactation is a period at risk of mastitis. Our objective was to investigate the influence of vitamin E or plant extract in the diet during a mammary local inflammation (LPS infusion) in dairy cows. 36 Holstein cows, with 14 primiparous cows, were classified into 3 groups. The 11 cows of the control group were fed an unsupplemented diet, the 13 cows in the vitamin E group received 3,000 IU/d for 4 weeks before and 1,000 IU/d for 5 weeks after calving, and the 14 cows at the same time in the plant extract group received 10 g/d. Blood and milk samples were collected before, and 4, 9, 28 and 76 hours after LPS infusion. White blood cell and milk somatic cell count were determined with a cell counter. Milk Na⁺:K⁺ was measured using the ICP-OES method as an indicator of mammary epithelium integrity. Milk cytokines were determined by ELISA. Gene expression was evaluated in milk by RT-qPCR fluidigm method. Data were analysed using a mixed model including group, time. Our results showed in T4 after LPS challenge, the white blood cell count was lower than before the challenge, and the milk somatic cell count increased at the same time, but were not influenced by the supplementations. The mammary epithelium integrity was higher in T4 in vitamin E and plant extract group than the control group. Respectively, in T4 and T9, cytokine IL8 and IL1 β were higher in plant extract group than the other groups. The expression of genes related to extracellular matrix (MMP9) and milk production (CSN1S1, SCD) were more expressed in the plant extract group than the control group. Genes related to extracellular matrix (SPARC) and cell junction (OCLN) were more expressed in vitamin E and plant extract groups than the control group. Our results showed that vitamin E or plant extracts improve mammary epithelium integrity and immune response, suggesting that these dietary strategies could reduce mastitis symptoms of dairy cows.

Phenotypic variation of colostrum yield and its association with immunoglobulin G content in Simmental cows

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Quantity and quality of cows' colostrum are pivotal elements for newborn calves. Colostral immunoglobulin G (IgG) are transferred to neonates establishing the first immunity. In dual-purpose breeds, promptly providing good-quality colostrum is crucial to have good performing young stock, i.e. future dairy heifers and beef animals. We disclosed the phenotypic factors affecting colostrum yield (CY, L) and IgG and estimated their correlation in Italian Simmental cows. 165 samples of first milking (< 6 h from calving) colostrum were collected and CY registered. Cows' parity ranged from 1 to 11, and 3 seasons were covered (summer, autumn, or winter). IgG was determined via near-infrared spectroscopy using self-built models installed in the DS2500 (FOSS Electric A/S, Denmark). Two linear mixed models were used for the ANOVA, one for CY and one for IgG. CY averaged 4.93 ± 2.33 L, with a mean IgG of 104.26 ± 39.02 g/L. Pearson's correlation between them was -0.15 ($P < 0.05$). CY was not significantly affected by parity, season of calving, type of farm (single- or multi-breed), interaction of parity and season. There was a significant difference between IgG content in cows belonging to the first two parity orders (lowest IgG) and the subsequent ones. IgG did not differ among calving seasons, farm type, or level of CY. Results indicate that CY does not change across parities and – given that the correlation between CY and IgG is weakly negative – there is no strong dilution effect in this breed whose official index currently include both milk and meat traits.

Plant extracts to optimize rumen fermentation from artificially reared lambs

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Artificially reared ruminants are sensitive to the weaning process given their slow rumen functional development. This in vitro aims to investigate the effects of dietary addition of plant extracts to optimize the rumen fermentation pattern using rumen inoculum from artificially reared lambs. A batch culture incubation was conducted for 24h using a high concentrate diet (92%), with incubation pH adjusted to 5.74. A dose-response set-up was planned with four doses (0, 0.1, 0.3, and 0.6 g/L) of six commercial feed additives: three essential oils blends (Agolin Ruminant and Biopracid, RumiG), allicin (Zooalium), saponins (Silvafeed MultiSap) and tannins (Silvafeed By-Pro). No significant differences were observed in rumen fluid pH and total volatile fatty acid concentration. All studied feed additives, except saponins, linearly decreased gas production during the late fermentation, whereas Agolin accelerated the gas production during the early fermentation. Supplementation with tannins, saponins and Biopracid promoted a linear decrease in the rumen ammonia concentration. Moreover, the essential oil blend RumiG was able to linearly increase butyrate molar proportion and increase the acetate to propionate ratio at its intermediate doses. These results suggest that the dietary supplementation with the tested plant extracts has minor effects on the rumen microbial fermentation during the post-weaning process of artificially reared lambs. The low rumen microbial development and absence of rumen protozoa, and the use of high concentrated feeds which lead to low rumen pH could explain the difficulty to modulate this microbial ecosystem. Further studies are needed with longer incubation times in order to allow the rumen microbiota to be adapted to the feed additive.

Characterizing rumen fermentation profiles associated with early lactation feed efficiency at different levels of milk production in transition dairy cows

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This study aimed to evaluate the effects of feed efficiency (FE) at two levels of milk yield (MY) during early lactation on rumen fermentation profile. Thirty-five multiparous Holstein cows were monitored from -35 to 60DIM. From -35 to -22 and -21 to 0 DIM, cows were fed far-off and close-up diets, respectively. From calving to 30 and 31 to 60 DIM, cows received fresh cow and post-fresh diets, respectively. Cows were retrospectively classified into 4 groups based on MY and FE: High MY and high FE (HH, n=11), high MY and low FE (HL, n=7), low MY and high FE (LH, n=7), and low MY and low FE (LL, n=10). The FE was calculated based on individual averages of DMI and MY. Spot rumen fluid samples were taken at 3h intervals to represent a 24h period at -14 and 14 DIM. From 1 to 11DIM, blood BHB was measured every other day. Data were analyzed with the PROC MIXED of SAS. A treatment effect (P=0.04) was observed for diurnal rumen pH at 14 DIM, where HH cows had greater (P≤0.03) rumen pH compared with HL, LH, and LL (6.8, 6.4, 6.4, and 6.4, respectively). A treatment effect (P=0.02) was observed in blood BHB concentrations, where greater (P≤0.01) BHB was observed in HH cows compared with HL and LL, and tended (P=0.08) to be greater than LH. Our results suggest that HH cows have a better ruminal adaptation to a high-energy diet after calving. Even though high blood BHB levels were predominant in HH cows, this did not impair MY or FE.

Session 49

Poster 30

Effect of a citrus-based sensory feed additive on heat-stressed dairy cows

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Climate change makes strategies for adaptation and mitigation of heat stress necessary to preserve livestock performances and health. The objective of this experiment was to evaluate the effect of a citrus-based feed additive (CE, citrus extract derived from *Citrus sinensis*) on performance of dairy cows suffering from heat stress. Primiparous early lactation Holstein cows (DIM 20) were enrolled in this study and were assigned to one of two treatment diets: 1) control TMR (CTRL; n=168) or 2) control TMR with 250 ppm of CE (VeO®; Phodé, Terssac, France) (CE; n=168). The experimental design included 3 successive periods of 40 days each. Cows received the CE during the second period. Milk yield, feed intake and rumination time were recorded daily as well as the ambient temperature (T) and relative humidity (RH). The level of heat stress was estimated by calculating the daily thermal humidity index (THI) : $THI = (1,8 \times T + 32) - (0,55 - 0,0055 \times RH) \times (1,8 \times T - 26)$. Values were classified into three categories : no heat stress (THI<68), light heat stress (68≤THI≤71) and moderate heat stress (71≤THI≤79). Data were analysed by ANOVA with treatment and THI category as fixed factors and the days in milk as covariate (General Linear Model, JAMOVI 2.3.26). DMI and milk production declined with increasing THI (P<0.05; R² = 0.06 and 0.29 respectively). DMI was numerically higher when the diet contained the citrus extract (CE 14.6 vs CTRL 14.1 kg/d; ns). Rumination time increased by 3.8% (CE 467 vs CTRL 485 min/d; P<0.001). Milk production was significantly reduced as THI intensified during CTRL period. Milk yield was higher with CE during light (CE 24.2 vs CTRL 22.8 kg/d; P<0.01) and moderate heat stress (CE 23.5 vs CTRL 21.9 kg/d; P<0.05). By modulating stress response, the citrus-based feed additive supported productive behaviours and milk yield under conditions of thermal stress.

MonoGutHealth: An Interdisciplinary approach for enhancing livestock resilience through nutritional strategies

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MonoGutHealth (MGH), a European Training Network project, targeted the complex challenges in pig and poultry production with an interdisciplinary strategy. The consortium leveraged diverse expertise in nutrition, feed chemistry, molecular biology, and more to foster disease-resilient livestock. Focused on training 11 PhD students with innovative methodologies, MGH emphasizes nutritional strategies for optimal gastrointestinal development and microbial colonization, aiming to bolster animal resilience. The research undertaken addressed early life interventions to reduce production diseases, advocating for sustainable and animal-friendly practices in producing pork, chicken meat, and eggs. By examining the phenotypic and metabolic profiles of underperforming livestock and identifying effective nutritional interventions, MGH strived for improvements in survival, growth, and welfare, testing these approaches under challenging conditions to ensure disease resilience. Complementing its scientific goals, MonoGutHealth offered comprehensive training to equip the students with essential skills and knowledge for their future professional paths. Through effective dissemination and communication, it seeks to inform stakeholders, policymakers, and consumers about its innovative approaches and achievements, contributing to sustainable livestock production. Funded by Marie Skłodowska-Curie grant (agreement no. 955374)

Differences between low and high body weight broilers and strategies for catch up growth of low performing chickens

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Despite genetic selection and advancements in farm practices, variability in body weight (BW) within flocks persists, posing economic challenges. We therefore aimed to identify factors influencing intra-flock BW variability in broilers and develop catch-up growth strategies for underperforming chickens. The first study investigated gut-related factors in BW variation, revealing reduced feed intake and weight gain, unfavorable histology traits, and higher pathogenic bacteria in low BW (LBW) chickens, while more growth-promoting bacteria in high BW (HBW) counterparts. Transcriptomic analysis showed higher gene expressions related to gut barrier function and nutrient transporters in HBW chickens, with lower expression of immune function and oxidation genes. Building on these findings, the second study investigated the effects of in ovo sodium butyrate (SB) injection (0, 0.1, 0.3, and 0.5% dose) on the 12th day of incubation, examining its impact on the growth performance, intestinal health, and gut microbiota of chickens with varying hatch weight (HW). Results revealed growth promoting impact of SB, with 0.3% SB showing higher chick performance. Interaction between HW and SB revealed that LHW chicks receiving 0.3% SB had improved BW and intestinal parameters compared to LHW and HHW controls but similar to HHW chicks with 0.3% SB. The third study examined the potential of dietary physical structures (coarse corn, oat hulls, or their combination) from day 7 post-hatch onward in enhancing gut development and performance in LBW broilers. All dietary treatments improved BW, gizzard weight and microbiota composition in LBW groups, with reduced gut permeability compared to LBW control fed standard feed. In conclusion, these studies elucidate factors influencing intra-flock BW variability and propose strategies, such as in ovo SB injection and dietary structural components during neonatal and post-hatching stages respectively, to optimize the performance of underperforming broilers and ensure flock uniformity during production. Funded by Marie Skłodowska-Curie grant (agreement no. 955374).

Effects of in-ovo stimulation on gut health and production of broiler chickens

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In the quest for alternatives to antibiotic use in broiler production, the effects of in-ovo stimulation with different bioactive substances on gut health and production parameters were studied. Prebiotic galacto-oligosaccharides (GOS), two probiotics (*Lactiplantibacillus plantarum* (LP) and *Leuconostoc mesenteroides* (LM)) and a prophybiotic (*Leuconostoc mesenteroides* + garlic extract (LM_G)) was used for the in-ovo stimulation on day 12 of incubation of ROS308 broiler chicken eggs. The treatments did not impair the hatchability, chick length and chick quality compared to the positive control (injected with physiological saline) whereas the chick weight was increased by the treatments at hatch and until 2 weeks of age. The meat quantity and quality were not adversely affected as a result of the in-ovo treatments when compared to the control. The histomorphometry in the ceca indicated an increased crypt depth in LM_G and LM treated chickens while the morphometry of the small intestine and ceca were similar between the groups. Moreover, the effectiveness of this approach was tested in response to a *Campylobacter* infection. The impact of the treatments on the gut microbiome and gene expression in immune related tissues of infected and non-infected chickens will be presented. Funded by Marie Skłodowska-Curie grant (agreement no. 955374).

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Theatre 4

Transcriptional and metabolic responses in chickens infected with mixed parasite species

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The performance of chickens can be impaired by nematodes *Ascaridia galli* and *Heterakis gallinarum* and protozoan *Histomonas meleagridis*. This study explores transcriptional and metabolic responses to mixed parasite infections using dataset S1 & S2. S1 contains liver transcriptomic profiles of 30 male birds examined at 2 weeks-post-infection (wpi) with mixed parasites. The birds belong to the genotypes (Ross-308 R, Lohmann Brown Plus LB, and Lohmann Dual-LD). S2 contains plasma metabolome profiles from laying hens (LB, N=108) examined at 2, 4, 6, 10, 14, and 18 wpi. Within-genotype analysis in S1 revealed 1438 differentially expressed genes (DEGs) in LD, while 199 and 17 DEGs were expressed in R and LB, respectively. Upregulated genes were mostly involved immune processes in R and LD but not in LB. Downregulated genes in LD and R were involved in metabolic pathways. Multivariate data analysis in S2 showed significant alterations in 20 metabolites at wpi 2, 6, and 10. Pathway analysis suggested increase in metabolic pathways, particularly amino acids pathways. The data suggest that elevated metabolic activity represents the metabolic cost of defence against parasitic infections. Changes in the hepatic transcriptome reveal immune-related activities dominating in livers of infected chicken, indicating a switch in hepatic functions during mixed parasitic infections. This project was funded by EU-Horizon 2020, grant agreement No 955374. NMR data were generated at FOODHAY, AU.

Monoguthealth in Denmark: Early interventions in pigs and broilers to improve their health and welfare
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Aiming at improving animal health and welfare, and reducing antimicrobial resistance, supplementation of plant-based antibacterials for broilers and piglets, and interventions during the incubation phase of broilers were investigated. Project-I studied the use of plant-based materials against post-weaning diarrhea in pigs and necrotic enteritis in broilers. Garlic combined with apple pomace or blackcurrant in the feed of weaners effectively reduced ETEC-caused postweaning diarrhea and positively influenced the gut microbiota. In broilers, the combination of palm kernel oil and clove essential oil acted synergistically *in vitro* against *C. perfringens* and effectively reduced intestinal lesions and bacterial loads of broilers under a necrotic enteritis challenge. Project-II addressed compromised welfare and mobility of broilers at the end of the growth phase by testing the influence of temperature during embryonic days 4–7 of incubation on locomotion. Temperatures of 36.5°C, 38.5°C, and 39.0°C were compared to a control of 37.5°C. Early incubation temperature influenced the muscle metabolism, with breast- and leg muscles responding differently. These changes did not subsequently affect walking ability. Bone strength, performance, and meat quality were unaffected. Concluding, while combinations of antibacterial plants and related products show promise in reducing the usage of conventional antimicrobials in piglets and broilers, more fine-tuning of the incubation program is needed to ultimately improve broiler health and welfare in production. Funded by Marie Skłodowska-Curie grant (agreement no. 955374).

Session 50

Theatre 6

Pre- and post-weaning liquid feeding of pigs

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Increasing pre-weaning feed intake of pigs by offering a liquid creep diet can increase intestinal maturity and weaning weight. Providing supplementary milk/liquid feed immediately post-weaning (PW) can increase average daily feed intake (ADFI) and average daily gain (ADG) during supplementation and may increase lifetime growth. Two pre- and two post-weaning experiments were conducted where liquid milk replacer (LMR) and a mixture of LMR and starter diet (LMR+S) were provided as creep feed and supplementary feed, respectively. In Experiment 1, providing LMR+S to suckling piglets tended to increase PW ileal villus height. In Experiment 2, LMR+S-fed piglets were 0.6kg heavier at weaning and had less pre-weaning mortality than pigs fed dry pelleted starter (DPS) diet. However, piglet growth PW was not increased by feeding LMR or LMR+S. Supplementing LMR to weaned pigs for 10 days increased ADFI by up to 85% and ADG by up to 94% during supplementation and body weight at day (d)10 PW by up to 21%. Milk-supplemented pigs had increased intestinal villus height and brush border membrane enzyme activity, decreased jejunal pro-inflammatory cytokine (IL17, IL18, IL22) expression at d7PW and increased faecal abundance of Rikenellaceae and Oscillaspiraceae at d10PW. Additionally, carcass weight at slaughter was increased by ~2.6 – 3.2kg in two experiments where LMR was fed for 10 days PW. Overall, liquid feeding pre-weaning can increase weaning weight and reduce pre-weaning mortality but had no effect on residual PW growth. Feeding supplemental milk replacer for 10 days PW can increase immediate PW ADFI and ADG and carcass weight at slaughter. However, milk replacer is expensive and its use must be economically justified. Funded by Marie Skłodowska-Curie grant (agreement no. 955374).

Glutamine supplementation during the suckling period and its influence on piglet growth and intestinal metabolism

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The suckling period is a critical developmental window, which influences the growth and health of piglets during subsequent life phases. Glutamine (Gln) supplementation is a dietary intervention that has been shown to improve the growth of suckling piglets. However, the mechanisms behind the improved growth remain poorly understood. In vitro studies report that Gln is the primary energy source for enterocytes of the small intestine (SI). In post-weaned piglets Gln supplementation is associated with improved intestinal health. However, the post-weaning physiological state is very different to that of suckling. Therefore, we investigated morphological and functional changes in the SI during the suckling period, in response to Gln supplementation. We have previously reported that the improved growth of low birthweight (L) suckling piglets supplemented with Gln was barely associated with changes in SI morphology, development, amino acid (AA) concentrations or mRNA abundance of genes involved in AA transport, metabolism, and glutathione synthesis compared to Alanine-supplemented controls. We are currently investigating SI metabolism using glucose (¹³C6-Glc) and Gln (¹³C5-Gln) stable isotope tracers, assessing SI mass and chyme AA content, of L suckling piglets supplemented with Gln, compared to water-supplemented controls. Results from our current and previous studies will be discussed in the context of what has been reported by other authors as will the suitability of Gln as a supplement to improve suckling piglet growth. Funded by Marie Skłodowska-Curie grant (agreement no. 955374); Deutsche Forschungsgemeinschaft (DFG): Q-PIG, ME 1420/10-1, ZE 322/4-1.

Effect of creep feeding on pre-weaning growth of suckling pigs

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Providing creep feed (CF) to suckling pigs can increase weaning weight (WW) and pre-weaning growth. The aim was to determine the effect of creep feeding on pre-weaning piglet growth, and if classifying piglets as eaters (E) or non-eaters (NE) of CF influences pre-weaning growth. Forty-five litters were randomly assigned to the following treatments; 1. No creep feed provided (CON; N=12 litters/treatment) and 2. Pelleted CF (16.25 MJ/Kg DE, 1.65% Lys) provided from day (d) 12 to weaning at d28±0.2 (CF; N=33 litters/treatment). Chromic oxide was added (1%) to CF from d16-25, and faecal swabs were collected from all pigs offered CF on d18, 21, and 25. Pigs were classified as E if they had 2 or more green swabs. Individual piglet body weight was recorded on d12, 19, and 28, and used to calculate average daily gain (ADG). Creep feed disappearance was recorded between pig weighings. Data were analysed using PROC MIXED (SASv9.4). Average CF disappearance was 227 g/pig during lactation. Within the CF litters, 27.6% and 72.4% of the pigs were classified as E and NE, respectively. On d12, pigs subsequently classified as E were lighter than NE (P<0.05) which in turn were lighter than CON (P<0.05); however, E and NE were heavier than CON on d19 and 28 (P<0.05). From d12-19, ADG was not affected by treatment (P>0.05); however, from d19-28 E and NE had a higher ADG than CON (P<0.05). In conclusion, creep feeding increased pre-weaning growth and WW. Furthermore, pigs classified as eaters of creep feed were found to have been lighter at commencement of creep feeding than those classified as non-eaters.

Feeding a gestation diet to sows in early lactation and liquid creep feeding of suckling pigs

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Feeding a gestation diet to sows in early lactation and liquid creep feeding can increase sow and piglet feed intake, respectively, leading to increased pig weaning weight (ww) and post-weaning (pw) growth. The objective was to determine the effect of feeding sows a gestation diet for the first 5 days of lactation and liquid creep feeding suckling pigs on pre- and pw growth in pigs. The experiment was a 2×2 factorial arrangement with 50 sows, that combined sow feeding (lactation diet throughout lactation or gestation diet for the first 5 days of lactation, followed by lactation diet) and creep feeding from day (d) 5 after birth (dry pelleted starter diet or liquid mixture of milk and starter diet). All sows were limit-fed a gestation diet until farrowing. Sow feed intake, weight and back-fat depth and piglet weight and creep feed dry matter disappearance during lactation and piglet growth to d14 pw were recorded. Sow milk was collected on d5 of lactation. Data were analysed using PROC MIXED (SAS v9.4). There was no sow x piglet treatment interaction for any parameter. Feeding the gestation diet in early lactation did not influence sow feed intake, back-fat depth or weight loss during lactation but reduced milk solids and fat % (P<0.05), tended to reduce ww (P=0.06) and reduced pw average daily gain (P<0.05). Creep feed disappearance was higher for dry-fed compared to liquid-fed litters but there was no effect of creep feeding on pre- or pw pig growth. In conclusion, feeding a gestation diet in early lactation did not increase lactation feed intake in sows and liquid creep feeding did not increase feed intake and growth in pigs compared to dry creep feeding.

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Poster 10

Maternal and/or direct feeding of organic acid-preserved grain improves pig lifetime growth performance

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Manipulating sow nutrition and incorporating organic acids (OA) into pig diets can improve pig health and performance post-weaning. This study evaluated the effects of including cereal grains preserved with an OA mould inhibitor (650 g/kg propionic acid) on the production performance of sows and their offspring. The 80 sows (Large White x Landrace) were blocked according to parity, body weight, and backfat on d 100 of gestation and assigned to one of two diets throughout lactation. On d 10 postpartum, the sow treatments were further divided. Their litters were assigned to one of two diets in a 2 x 2 factorial design (n = 20), resulting in 4 treatments: (T1) DSDP (dried grain sow diet + dried grain pig diet); (T2) DSPP (DS + preserved grain pig diet); (T3) PSDP (preserved sow diet + DP); and (T4) PSPP. The piglets remained on their respective diets post-weaning and were monitored until slaughter, at d 168. Sows offered the OA-preserved grain had improved apparent total tract digestibility of nitrogen and gross energy (P <0.05). There was no effect of diet on sow reproductive performance (gestation length, liveborn, milk production, and wean to oestrus interval), body weight loss, backfat loss, or feed intake during lactation (P > 0.05). There was no effect of treatment on pre-weaning piglet performance (P >0.05). However, a direct feed effect was observed between d 26 and 168 postpartum where pigs offered the OA preserved grain had improved daily gain, feed efficiency, final body weight and carcass weight at slaughter compared to dried grain (P<0.05). Pigs weaned from OA-preserved grain-fed sows had improved FCR during the finisher stage (P <0.05). In conclusion, OA-preserved grain improved the lifetime growth performance of pigs.

Organic acid-preserved grain improves growth performance and gut health of post-weaned pigs

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The objective of this study was to investigate the effect of preserving cereal grains with an organic acid (OA) mould inhibitor (650 g/kg propionic acid) compared to conventional grain drying on pig health and performance post-weaning (PW). A 2 x 2 factorial design assessed the effect of grain preservation method on growth, diarrhoea scores, nutrient digestibility, and gut microbial populations, with or without zinc oxide (ZnO) supplementation. The 192 pigs (n=16) were assigned to either: (T1) dried grain diet, (T2) preserved grain diet, (T3) dried grain diet + ZnO or (T4) preserved grain diet + ZnO for 35 days. Significant interactions between preservation method and ZnO inclusion were observed on feed intake (ADFI), daily gain (ADG), and final body weight (BW). The OA-preserved grain improved ADG (P < 0.01) and BW (P < 0.01) compared to the dried grain. ZnO supplementation increased ADFI (P < 0.01) and BW (P < 0.05) in the dried grain diet, however, it did not affect ADFI (P > 0.05), and reduced ADG and BW in the preserved grain diet. ZnO inclusion reduced faecal scores and diarrhoea incidence compared to non-ZnO supplemented pigs (P < 0.001). Pigs offered the preserved grain diet had reduced faecal scores and diarrhoea incidence compared to the dried grain diet (P < 0.05). The preserved grain group had an improved coefficient of apparent ileal digestibility of nitrogen and gross energy (P < 0.01), lower levels of colonic branched-chain VFAs (P < 0.05), and favourable shifts in gut microbial populations, including reduced *Streptococcus* in the ileum (P < 0.001) and an increase in *Faecalibacterium* in the colon (P < 0.05). In conclusion, organic acid-preserved grain improved nutrient digestibility and gut health, thus enhancing growth performance PW.

Session 50

Poster 12

Effects of multi-strain probiotic supplementation on microbiota and performance in pigs

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The purpose of this study was to evaluate the effect of multi-strain probiotic (MSP) supplements on feed growth performance and intestinal microbiota composition. MSP was the combination of 1.1×10^9 CFU/g *Lactobacillus ruteri* CACC607, *Pediococcus pentosaceus* CACC616, *L. dextrinicus* CACC889, *L. pentosus* CACC891, and 7.6×10^4 CFU/g of *Saccharomyces cerevisiae* CACC699. A total of 37 piglets with an initial average body weight of 7.40kg, were divided into two treatment groups (control = 20, MSP = 17). The production parameters like average daily feed intake, average daily gain, and feed conversion ratio were calculated based on the data measured daily. The MSP group showed a lower feed conversion ratio than the control group. Furthermore, the MSP group enhanced the production and health-related genera: *Dorea*, *Faecalibacterium*, *Lactobacillus*, *Blautia*, and *Butyrivibrio*. Notably, the MSP group showed reduced digestibility-related odor compared to the control group (P < 0.05). Based on the results, MSP can be considered as the functional probiotic preparation for feed supplements and odor reduction. This research was funded by the Science and Technology Project Opens the Future of the Region (Project No. 1711177233), the Innopolis Foundation, and the Korea Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry (IPET) through the Agri-Food Export Enhancement Technology Development Program, funded by the Ministry of Agriculture, Food and Rural Affairs (MAFRA) (RS-2023-00234143).

Effects of *Lactobacillus pentosus* supplementation on growth performance and short-chain fatty acids in feces of growing to finishing pigs

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This study was conducted to investigate the effects of supplementing *Lactobacillus pentosus* in powder form (LP) on growth performance and short-chain fatty acids (SCFA) levels in feces of growing to finishing pigs. A total of 48 growing pigs [(Landrace×Yorkshire)×Duroc] (2 pigs/pen, 6 pens/treatment) with an initial average body weight (BW) of 33 kg were used for 75 days. The dietary treatments included a corn-based diet with four levels of LP supplementation: 0, 0.1, 0.2 or 0.3 % LP. Data were analyzed using 1-way ANOVA and orthogonal polynomial contrast. Increasing LP supplementation resulted in a linear increase ($P<0.05$) in average daily gain (ADG) and feed efficiency. In the overall period, the group supplemented with 0.3% LP exhibited the highest final BW, significantly higher ($P<0.05$) than that of the non-supplemented group. As for feed efficiency, LP supplemented groups had the highest ($P<0.05$) final BW than the non-supplemented group. Moreover, increasing the LP supplementation showed ($P<0.05$) linear effect in average daily gain (ADG) and feed efficiency. At the end of the growing period, 0.1% and 0.3% LP supplementation increased ($P<0.05$) acetic acid and total SCFAs concentration in the feces compared to non-supplementation. Moreover, isovaleric acid and total SCFAs concentrations in the feces linearly increased ($P<0.05$) as LP supplementation increased. The study's findings indicate that supplementation with LP positively impacts growth performance and enhances SCFA levels in the feces of growing-finishing pigs.

Session 50

Poster 14

Effects of valerate glycerides in vitro using porcine cell culture models

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Short chain fatty acids are understood to exert biological activities, however, there is limited research regarding impacts of valerate on host physiology. The current experiments aimed to evaluate anti-inflammatory capacity and effects on intestinal barrier integrity of valerate glycerides using in vitro porcine models. All tested doses were confirmed non-cytotoxic by MTT assay. Porcine alveolar macrophages (PAMs) were harvested via bronchoalveolar lavage from piglets ($n = 6$) and seeded at 6×10^5 cells/well for culture overnight. Then, cells were treated with 0, 250, 500, or 1000 $\mu\text{g}/\text{mL}$ valerate glycerides with or without lipopolysaccharide (LPS) challenge (1 or 0 $\mu\text{g}/\text{mL}$). After 24 h, supernatants were collected and concentration of pro-inflammatory cytokine, TNF- α was analyzed by ELISA. For non-challenged PAMs, the tested doses had no effect on TNF- α secretion. Treatment with 500 $\mu\text{g}/\text{mL}$ valerate glycerides significantly ($P < 0.05$) reduced TNF- α secretion in PAMs under LPS-challenge conditions. In the second experiment, several passages of IPEC-J2 cell line were seeded ($n = 3 - 4$) at 5×10^5 cells/mL in transmembrane inserts. After differentiation, cells were treated with valerate glycerides (0, 100, 500, or 1000 $\mu\text{g}/\text{mL}$) to assess the integrity of IPEC-J2 monolayers at several timepoints (0, 24, 48, 72 h post-treatment) by measuring transepithelial electrical resistance (TEER, Ωcm^2). Compared with control (0 $\mu\text{g}/\text{mL}$), treatment with 100 or 500 $\mu\text{g}/\text{mL}$ had no impact on TEER. Treatment with 1000 $\mu\text{g}/\text{mL}$ significantly decreased TEER compared with control. Further investigation is warranted to understand interactions with valerate glycerides and porcine cells in vitro.

Evaluation of the effects of a standardized citrus extract on the intestinal microbiota and welfare of peripartum sows under commercial breeding conditions

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Farrowing is a critical period for sows, which is often accompanied by digestive disorders with negative consequences for animal health and welfare. To manage these situations, intestinal microbiota modulation could be a good solution. In this context, the effects of a standardized citrus extract (SNCE) on peripartum sows and their offspring were evaluated. Fifty sows were divided into 2 groups: a control (CTL) group fed a standard diet and an SNCE group supplemented with 2500 ppm of SNCE, from 10 days before farrowing until 5 days after. Performances and transit resumption were monitored. At the end of the supplementation phase, 8 sows per group were randomly selected for microbiota analysis. Results showed better feed intake for sows from SNCE group (8,540 g/d) compared to CTL group (7,937 g/d, $p < 0.01$). Moreover, the interval between farrowing and first defecation was also reduced in the SNCE group (1.35 Vs 1.88 in CTL group, $p < 0.05$), which indicates a better and faster transit resumption after farrowing. Sows' fecal microbiota analysis revealed two different bacterial ecosystems in the 2 groups. Regarding piglet performances, SNCE supplementation increased their weight gain between 24h and 7 days after farrowing (1.35 kg), compared to CTL group (0.93 kg, $p = 0.02$). In conclusion, these results show that SNCE supplementation improves the welfare of sows around farrowing by reducing constipation. This positive effect is correlated with sows' microbiota modulation, which could partly explain the effects observed in sows and piglets.

Session 50

Poster 16

Effects of maternal probiotic, and/or piglet dietary tryptophan supplementation pre-weaning, on sow and offspring performance

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Probiotics and prebiotics hold promise for enhancing animal health and performance by positively modulating the microbiome. Maternal probiotic supplementation benefits both the sow and her offspring as it leads to transmission to the offspring. Amino acids, particularly tryptophan (Trp), have emerged as innovative prebiotics. Combining maternal probiotic supplementation with direct offspring prebiotic supplementation could synergistically enhance postnatal pig microbiome establishment, improving growth and performance. A 2x3 study was conducted to investigate the effects of maternal probiotic supplementation, and/or increased piglet Trp supplementation, on sow and offspring performance pre-weaning. On D85 of gestation, 48 sows were grouped by parity and backfat and assigned to one of two groups: 1) basal or 2) basal + *Bacillus subtilis* and *Bacillus amyloliquefaciens*. On D8 postfarrowing, litters were further divided and assigned to one of 3 creep diets resulting in 6 groups (n=8 litters/group): T1) B17 (basal sow diet+0.17 Trp:lysine creep); T2) B21 (B+0.21 Trp:lysine creep); T3) B25 (B+0.25 Trp:lysine creep); T4) P17 (probiotic sow diet+0.17 Trp:lysine creep); T5) P21 (P+0.21 Trp:lysine creep); T6) P25 (P+0.25 Trp:lysine creep). Diet had no effect on liveborn, birthweight, weaning weight, piglet average daily gain, or sow backfat loss ($P > 0.05$). There was a tendency for reduced lactation feed intake in probiotic sows while their litters tended to have increased creep intake ($P < 0.1$). Maternal probiotic x 0.25 Trp:lysine creep tended to increase the number of pigs weaned per litter and reduce mortality compared to other groups ($P < 0.1$). In conclusion, there was minimal effect of maternal probiotic supplementation or level of Trp in the creep on sow or offspring performance.

Genetic progress vs animal welfare?

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In this joint challenge session of Animal Health and Animal Welfare – Genetics and FABRE TP, we will discuss Animal Breeding and Genetics in relation to Animal Welfare in future sustainable livestock systems. We invited experts to have constructive debates on the role of animal genetics on the current welfare of farm animals, and how animal welfare can be enhanced and considered in future breeding programs. The regulatory framework, ethics, and societal context of food production will be considered in this discussion with all the necessary and relevant stakeholders. Code EFABAR and the concept of balanced breeding alongside technological advances and the evolution of welfare and behaviour sciences will be also part of the conversation.

Session 51

Theatre 2

Reversing the detrimental impact of genetic selection on farm animal welfare

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Animal welfare policy rightly gives considerable attention to the physical conditions in which farm animals are kept. Much less consideration is given to the impact of genetic selection for enhanced productivity on animal welfare. The use of genetic technologies is likely to receive increased attention as English law permitting the use in farming of gene edited animals is likely to be in force shortly and the EU may introduce similar legislation. Modern meat chickens have been bred to grow over twice as quickly as 60 years ago, causing millions in the EU to suffer from leg disorders each year. Hens have been bred to lay over 300 eggs a year. They have to draw on their own bone calcium to form eggshells making them susceptible to bone fractures. Today's dairy cows have been bred to produce ten times more milk each year than they would naturally produce. Selective breeding is indirectly pushing animal agriculture towards greater intensification. For example, the production of large litters of piglets leads to increased competition for teats and so to injuries to the sow and litter-mates. This results in the use of teeth clipping to prevent injuries. Breeding for excessive litter size also leads to the use of artificial rearing systems and nurse sows to foster surplus piglets. EU legislation provides: "No animal shall be kept for farming purposes unless it can reasonably be expected, on the basis of its genotype or phenotype, that it can be kept without detrimental effect on its health or welfare". This is good in principle but is widely ignored in practice. Legislators and the breeding and livestock sectors must work together to end the suffering caused by selective breeding and to ensure that gene editing and other novel breeding technologies do not have adverse impacts on animal health.

The evolution of dairy cattle breeding objectives

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Purpose of this overview is to present the evolution of breeding goals in dairy cattle, aimed at a more sustainable future. Since the 1970s, selection objectives and the actors changed, including the technologies. The dairy production system market changed in response to social needs and industry demands. Modern animal breeding moved away from direct selection solely based on milk yield. Today the goal is to address farmers' needs and engage a broader range of objectives and stakeholders. The changes reflect a growing social awareness of animal welfare and the trilogy of sustainability (social, environment and economic). Today, we face a more sustainable selection program, with a focus on holistic animal husbandry. This approach benefits animals, breeders, consumers and society. From the 1970s to the present day, we have witnessed ongoing advancements in data collection based on precision farming technologies. Precision of recording phenotypes remain the fundamental basis for genetic evaluation model and genetic progress. Selection of high-yield cows has led to efficient animals, nonetheless the environmental requirements were strong. Cows have limited resources available for supporting a new pregnancy, fighting infections, and dissipating excess heat. This challenge can be faced. Progress is made since fertility and longevity were included in breeding goals, resulting in a genetic progress for these traits. Similarly, efforts to select for improved resilience and heat tolerance are already implemented. The evolution of breeding goals is also incorporating environmental efficiency, addressing global warming, enhancing rumen efficiency and developing selection indices for improved holistic efficiency. Innovative approaches like the use of the community capitals framework, artificial intelligence combined with big data provided by sensor technologies, are improving animal welfare and minimizing environmental impact. Looking ahead, the challenge of selection lies in integrating multi-disciplinary approach to create a more sustainable and resilient food production system.

Session 51

Theatre 4

Predicting the agility of breeding programs to adapt to future societal demands

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Societal demands and evolving policies will cause significant transformations in future farming systems. Consequently, the requirements for animals kept in those systems may change, thereby influencing breeding goals. From the perspective of breeding programs, two key priorities emerge: firstly, more accurately predict potential changes in societal demands in the coming decade, and secondly, create the adaptability to swiftly respond to such changes. Moreover, predicting the effects of any changes in the breeding goal or breeding program on the results of the selection process is crucial. Conversely, for stakeholders such as farmers, retailers, and policymakers, who articulate specific requirements for animal production, it is important to understand the responsiveness of breeding programs. Thus, having insights into the response time of breeding programs is essential. In this context, the availability of a digital twin becomes highly relevant for breeding programs, offering a means to model and predict the impact of anticipated societal changes. This digital twin can be practically realized by mirroring the breeding program through a comprehensive stochastic simulation, a process facilitated by tools like the MoBPS package. During the conference, practical cases will be showcased, illustrating potential abrupt changes in breeding goals. The presentation will go into the subsequent effects on other traits and highlight the timeframe necessary to implement these changes.

Breeding goal scenarios to quantify consequences of EFSA recommendations on pig reproductive traits

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Productivity in pig production is increasing, among other factors through genetic change. There is societal and academic concern that, especially litter size is increasing to levels beyond the capacity of the sow in our current production environment. EFSA suggests husbandry changes (e.g. free farrowing) and changes in selection for a number of traits, e.g. birth weight (BW), uniformity in birthweight (LVAR), mothering ability (MAB), number of teats (NTE), and litter size (TNB). This abstract presents an attempt to objectify the discussion between welfare and economy and what can be realized through selection in what period of time. The MoBPS package was used to create a digital twin of a pig breeding program, simulating large numbers of individual animals at the SNP level, with phenotyping, reproduction, and especially index selection. The basic scenario was pedigree selection for finisher traits (daily gain, feed efficiency, and backfat) and reproduction traits (TNB, BW, LVAR, MAB, NTE). The use of genomic information increased the economic output with 30% (an internal test). An extreme scenario, zero economic value for TNB, resulted in a drop in litter size over the time period of 100 months. Zero value for mortality yielded a substantial increase in litter size. During the conference more nuanced scenarios will be presented and the discussion should be on the relevance of digital twins for the discussion between economy and welfare.

Do pig farms reconcile sow prolificacy, pre-weaning piglet survival and weaned litter size?

Typology and results of French herds

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A mean pre-weaning mortality rate of at least 20% remains a key issue in pig production for economic, technical and societal reasons. Negative impacts of large litters and poor adjustment to the number teats available are regularly blamed, with potentially higher risks associated with free-farrowing systems. The objective of this study was to analyse the high variability in herd results and identify farm profiles that had an optimum combination of weaning performance, piglet survival and litter sizes. Data were extracted from the French database “Technical Management of Sow Herds”, managed by IFIP. Mean results of 1013 farms in 2022 were used, along with their main characteristics (region, size, conventional vs. organic production) and some sow-housing characteristics (e.g. outdoor farrowing, free-farrowing, farrowing lift cages). Their results, 16.4 ± 1.3 total born, 15.2 ± 1.0 live born and 12.7 ± 1.0 weaned per litter, are associated with variable and increasing mortality rates: 22.3 ± 5.3 % and 15.8 ± 4.8 % mortality for total born and live born respectively. A herd typology was developed using hierarchical ascendant classification. Five distinct groups were identified that had contrasting increasing numbers of weaned piglets, with different characteristics and combinations of results. Analysis of these groups shows that despite unfavorable correlation between large litters and pre-weaning survival, high mortality rates can occur with all litter sizes. However, some farms were able to reconcile good weaning performance, large litter sizes and low pre-weaning mortality. These results highlight possible benefits of a multicriterial approach for setting farm targets, specially for alternative sow housing systems.

HenTrack: Improving breeding programs with superior behavior and welfare phenotypes

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While the transition to cage-free systems is positive for laying hen welfare, a major obstacle exists in that breeding paradigms are dependent on the use of small groups of hens that mirror cage systems. Compared to cages, cage-free housing systems (HS) provide a rich array of resources, including but not limited to ranging areas, litter for dustbathing, and elevated roosting positions. Beyond these resources, the social structure will also be drastically different where flock sizes can exceed 20,000 animals. It is well established that while hens will develop a social (pecking) order of dominance in small groups, groups above 70-80 animals do not adopt this social order. Under current breeding paradigms, genetic traits that perform well within small groups of hens and effectively deliver the high productivity of modern commercial operations must also perform well within the very different environment of HS. Unfortunately, this is often not the case, as many problems exist within HS. The widespread occurrence of these issues across Europe, which were early adopters of cage-free housing systems, proves the difficulty in resolving the problems and the limitations of non-genetic solutions (e.g. improved management). The Center for Proper Housing of Poultry and Rabbits (ZTHZ) has been leading an effort to assess individual behaviour within large groups of hens housed in commercial environments. Beginning with early work showing a bimodal distribution in laying hens' use of an outdoor range, more recent work has shown highly consistent movement and location patterns of animals both within the barn as well as in the use of outdoor areas. By combining the ability to quantify movement using non-invasive behavioral tracking with measures of health, our group is leading a large effort to identify behavioral markers of health in collaboration with two of the world's largest provider of genetics. The HenTrack project (<https://tinyurl.com/Avinew>) benefits from the provision of specialized parental crosses that link directly to each company's Pure Line stock, allowing identified traits to be implemented in subsequent breeding plans and maintain the goal of more robust hens better suited to HS that maintain expected health and productivity standards.

Session 52

Theatre 1

Agroecological assessments as an opportunity for more resilient and sustainable livestock systems

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The controversy around livestock farming systems reflects a complex interplay of environmental, ethical, health, economic, and cultural factors, making it a multifaceted and challenging issue to address. Solutions often involve a balance between meeting the nutritional needs of a growing population, ensuring economic stability for farmers, and minimizing the environmental and ethical impacts of livestock farming. Recently, several publications addressed the need for holistic assessments of agricultural systems with less focusing on livestock. Moreover, available tools differ in completeness and complexity. The Tool for Agroecology Performance Evaluation (TAPE), developed by the Food and Agriculture Organization of the United Nations (FAO) and multiple partners, provides an analytical framework for the multidimensional assessment of sustainability for agricultural and livestock systems. Here we provide insights into the development of the tool to better represent two environmental dimensions, namely biodiversity and the contribution to climate change. Simultaneously, we assess resilience to external stresses such as those caused by climate change. The tool allows farmers and other stakeholders to assess current farming systems and identify opportunities to optimize livestock systems in the future.

Effects of Supplementation with Tannins and Saponins Blend on Methane Emissions in High-Emitting Dairy Cows

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Tannins and saponins are plant secondary products utilized in ruminant farming, positively impacting ruminal fermentation and reducing methane emissions (CH₄). Moreover, literature highlights variations in CH₄ potential among cows, influenced by specific conditions that may increase or decrease daily CH₄. We aimed to (1) evaluate the CH₄ potential of a dairy herd; (2) test Silvafeed BX (0.08% DMI, TAN), a blend of hydrolysable and condensed tannins and saponins on high CH₄ emitters. A commercial herd (40 dairy cows) was tested for CH₄ emissions using a portable laser device. The common diet was based on alfalfa and wheat hays (47%) and concentrates (beetpulp 7, corn & sorghum flakes 32, soybean 10, molasses 1, supplements 3 % of DM). 5 Italian Friesian cows had higher CH₄ and were enrolled in the trial composed of baseline (7 d, P1), adaptation (14 d), and TAN (7 d, P2) periods. Records and sampling were performed on P1 and P2. CH₄ was scanned every 8 hours for 2 days in P1 & P2. Rumination time (RT) and reticular pH (r-pH) were measured using a rumination monitoring system and reticular boluses, respectively; dry matter intake (DMI) and milk yield (MY) were individually recorded. Rumen fluid VFA and ammonia, organic matter digestibility (OMD), and milk composition were determined. Energy-corrected milk (ECM) and dietary nitrogen retention (DNR) were calculated. A mixed model procedure was used for data analysis; TAN presence was used as a fixed effect. TAN addition led to reduced CH₄ (from 502 to 457g/d, P<.01) as well as CH₄/DMI (-11%, P<.01), CH₄/MY (-16%, P<.01) and CH₄/ECM (-14%, P<.01). Higher MY (+2.9 kg/d, P<.01) and milk protein (+0.19%, P<.01) were reported on P2, ECM (+1.8kg/d, P<.01) and DNR (+5%, P<.01) increased as consequence. DMI (26.2kg/d), RT (496min/d), r-pH (6.25), total VFA production (99mmol/L), rumen ammonia (5.59mg/dl) and OMD (66%OM) resulted not influenced. Obtained results showed that feeding Silvafeed BX in high CH₄ dairy cows produces a reduction of emissions, improved milk production and nitrogen utilization.

Relationship between environmental impact and on-farm dairy cow welfare in Parmigiano Reggiano farms

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Parmigiano Reggiano (PR), a very hard and long-ripened cheese, is one of the most significant dairy chains in Italy, under the regulation of a specific Consortium. This study aimed to assess the relationship between global warming (GWP, kg CO₂-eq) and acidification potential (AP, g SO₂-eq) and on-farm animal welfare in PR farms. Data derived from 243 farms (145±159 cows, 26±6 kg fat- and protein-corrected milk – FPCM/lactating cow/d). The GWP (global-related impact, per 1 kg FPCM) and AP (local-related one, per 1 m²) were estimated with Life Cycle Assessment. All farms underwent an assessment of animal welfare with IZSLER/CRenBA protocol, that encompassed various areas, including management (A), structure (B), and animal-based measure (C), utilizing a scoring system ranging from 0 to 100. Values of GWP, AP and A-B-C areas scores were analysed with Pearson's correlation analysis. The GWP resulted positively and significantly correlated with AP and negatively with A and B scores, although all with low r values (0.25, -0.37 and -0.23, respectively). AP resulted not correlated with A, B or C scores. Within welfare scores, A and B resulted highly correlated (r = 0.60). When considering only on-farm AP, no correlation was found with GWP, A, B or C scores. In conclusion, various aspects of a dairy farm's overall sustainability exhibit complex relationships. However, there is room for improvement in one aspect without worsening some others. Acknowledgements: Marco Berton was supported by project "Transition towards GREENing for LIVEstock farming systems". REACT EU PON 2014–2021. Ricerca e Innovazione, Asse IV, azione IV.6.

Modeling interactions within dairy farms using regular vine-copula models

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Developing mitigation strategies to reduce greenhouse gas emissions from farms is a major issue in the context of climate change. Nevertheless, sources of emissions are influenced by many interacting factors on the farms and in their environments. This raises the issue of how to adequately model the multiple interactions of on-farm and environmental factors to improve understanding of farm performances. To this end, we investigated regular vine copulas as a statistical modeling approach that enables mapping multivariate complex dependences using a graphical representation in a tree structure. The method was applied to a dataset of management practices, emissions and productivity of French dairy farms. When considering all farms, methane emissions depended on milk production via manure management, since milk production strategies influence both herd management and manure type. Dependences were also identified between extreme values of enteric methane emissions and the amount of digestible organic matter ingested, with a strong correlation. When considering farms as a function of milk production, while dependence between milk production and total dry matter ingested was the strongest for the most extensive farms, that between milk production and enteric methane emissions was the strongest for more intensive farms. We provided a new way to describe dependences among a set of variables of farms, even among their extreme values, increasing understanding of effects of practices depending on the farm context. Acknowledgments. We thank the French Livestock Institute for providing data and the Erasmus+ 2021-2027 program, Doctoral College of Brittany and Directorate for Higher Education, Sites and Europe as part of the program EIR-A for their financial support.

Global forecasted scenarios of THI and HLI anomalies

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Rising global temperatures are causing an increased risk of heat stress in livestock and a significant expansion of the geographical areas affected. In this study, anomalies of the Temperature-Humidity Index (THI) and Heat Load Index (HLI), two bioclimatic indicators, were used to assess the evolving global spatiotemporal patterns of heat exposure. The NEX-GDDP-CMIP6 dataset from NASA, along with the Community Earth System Model version 2 (CESM2) from the National Centre for Atmospheric Research (NCAR), were utilized for the analysis of comprehensive global data on air temperature, solar radiation, wind speed, and humidity at a high resolution of 0.25-degree grids. The THI and HLI were computed using climatological normal (CliNO) period data (1985-2014), as well as for future projections under various Shared Socioeconomic Pathways (SSPs) from 2026 to 2100 at 25-year intervals. Remarkable THI and HLI deviations were identified through the comparison of SSPs projection data against the CliNO, highlighting climate anomalies and providing insights into potential changes in heat stress risks. The analysis pointed out a trend of escalating anomalies, especially in the latter half of the century (2051-2075 and 2076-2100), with more noticeable effects under the high-emission scenario SSP5-RCP8.5. Projections for 2051-2075 within this scenario indicate a rise in average annual THI and HLI of over 4 units for regions such as North America, Europe, and India. Distribution of the anomalies was not uniform, and their magnitude was particularly pronounced in the tropics, subtropics, and some temperate regions. Results reported herein underscore the need for implementation of anticipatory adaptation strategies, which may help to preserve animal health and welfare and to ensure food security.

Measuring farm resilience attributes: The study of three small ruminant systems

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Agricultural systems need to be resilient to cope with challenges. Usually, resilience assessments do not focus on the system characteristics that contribute to building resilience, i.e. the resilience attributes. This is partly due to a lack of operational approaches to measure them. The aim of our research is to operationalise the measure of farm attributes. We considered 21 resilience attributes and developed 85 proxy indicators to measure attributes at the farm level. We applied the approach to three Spanish case studies: (i) meat sheep, (ii) dairy sheep, and (iii) dairy goats. Data were collected through 3 focus groups with local experts to assess the importance of the attributes in the case studies and through 144 surveys to farmers to measure the indicators. Based on these data, we calculated an overall resilience score. The scores obtained ranged from 25 to 75 out of a maximum score of 100. We found some strengths and weaknesses of each case study. Our research provides a tool that can be used by policy makers, farmers, banks, and other stakeholders to assess the status of resilience attributes and where improvement is possible.

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How resilience attributes contribute to specific resilience at farm level? The case of small ruminants in Spain
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The small ruminants farming systems (SRFS) with local breeds in Spain are in decline due to the challenges they face. The resilience attributes could help SRFS coping with these challenges. The aim of this work was to analyse how resilience attributes (n=21) modulate resilience to specific challenges (n=7) at farm level, as perceived by farmers. To this end, 144 farmers were surveyed in three case studies (dairy goat, dairy sheep, and meat sheep) in Spain. The survey enquired about the perceived resilience and the effect of challenge in a Lickert scale (1-7). In addition, resilience attributes score was developed based on indicators of farm characteristics and relations. Thus, an ordinal logistic regression was performed to analyse which resilience attributes would explain the farm' specific resilience to each challenge. Results show the effect of the challenge vary between cases. The most significant attributes are the family support and the exposition to disturbance in the dairy goat; family support, organization of the sector and the financial capital in dairy sheep; and time-space heterogeneity and functional diversity in meat sheep. Although, there are common points between cases, results show that depending on the case and the challenge resilience attributes influence differently the perceived specific resilience. These findings underscore it is necessary to understand the characteristics and structuration of livestock systems to identify areas requiring looking at for greater resilience.

Climate Change and Livestock Production: Revisiting Strategies to Ensure Sustainable Livestock Farming Systems

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The effects of climate change have negative impacts on global livestock production. Also, this sector contributes to global warming via greenhouse gas emissions. Ensuring farming systems can ameliorate the impacts of climate change while reducing the contribution of livestock to climate change should ensure sustainability. This could be attained by a multi-disciplinary approach utilizing molecular biology and bioinformatic developments. Using advanced ‘omics’ technologies like genomics, transcriptomics, metagenomics, epigenetics, and proteomics can lead to a deeper insight into the molecular mechanisms underlying climate resilience of livestock allowing the identification of several molecular markers for marker-assisted selection or genomic breeding programs. There is also rising interest in non-invasive approaches utilizing saliva, feces, urine, hair and milk to assess stress in livestock. Likewise, sensor-based approaches like rumen bolus, activity sensors, and infrared thermography have promising roles in assessing stress in livestock. Integrating the stress measures and climatic variables will allow modelling approaches to assess the resilience potential of breeds and individuals. Further, digitalization can aid towards ensuring climate-smart livestock production where stressed animals are identified quickly (eg. using sensors) or better resilient animals can be identified (eg. using ‘omics’), or the future performance of an individual could be predicted by modelling. Another vital component of climate-resilient livestock farming would be methane reduction achieved through management, genetic and nutrigenomic strategies. Thus, breeding for climate resilience and low methane emission while improving productivity is needed. Further, farmers need to adopt integrated farming systems, which can be more productive and reduce carbon footprints. Ensuring a resilient farming system involves diversified factors that can aid towards attaining climate-resilient sustainable farming.

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Remote monitoring of pasture and animals in extensive farming is functional to genetic improvement

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The IoT (Internet of Things) technology enables data collection and real-time analyses, which are less common in extensive farming situations. However, in these systems, it is also becoming increasingly important to collect information about feeding and animal behavior. Satellite data analysis on pastures provides grazing area information, enabling short-term farmer decisions to direct the pasture in areas with greater feed yield and quantity, and helping to reduce grazing short and long-term ecosystem impacts. In the Agritech project, a Machine Learning (ML) approach is under developing to estimate pasture yield and quantity. The estimation is performed using band and vegetation indexes retrieved from the Sentinel satellite, and climatic and altitude information. Grassland data were obtained from two different farms in Lazio, Italy, from June 2023 and still ongoing. More than 200 data from the field samples (eg, dry and fresh matter) and vegetation indexes (eg, NDVI) were used in the model. Simple correlation between satellite data and pasture features not exceed 0.45. The ML model identified a combination of bands, indices and climatological data that can provide better accuracy of pasture quality and quantity than using singular bands or indices as a feature. The preliminary results showed a greater importance of individual bands and indices than climatic data. For example, fiber, dry and fresh matter models obtained an R-squared value around 0.5; for protein the R-squared value reached 0.6. Nevertheless, an improvement of the model accuracy is expected due to the increasing of samples. This model provides a valuable tool for farmers, researchers and breeding associations. Furthermore, the combination of this information with other data (eg, animal sensors, videos), may support fine-tuning of genetic breeding programs based on identification and genetic characterization (“omics” technologies) of animals that differentially respond to environmental constraints.

Restoration of abandoned Alpine Summer Pastures: Insights into Vegetation, Soil Conditions, and Microbial Communities

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Alpine grasslands, recognized as High Nature Value Farmland due to their biodiversity, are crucial agro-ecosystems for extensive livestock systems. Within a project aimed at restoring a recently abandoned pasture located at 1900 of elevation in the northeastern Italian Alps, this study aims at assessing the pre-restoration interactions between vegetation, soil features and microbial communities and N cycle potential functions. Vegetation was analyzed with the Braun-Blanquet method during 2022, revealing three vegetation classes across ten sample areas. Topsoil samples from each area were examined to determine organic carbon content and pH. In addition, we used qPCR and sequencing to examine soil microbial communities. We used qPCR to quantify target genes involved in nitrogen cycle processes, evaluating their functional potentials. The targeted genes included *nifH* for nitrogen fixation, *amoA* for nitrification, *nirK*, *nirS*, and *nosZ* for denitrification. Sequencing was applied to characterize the microbial communities' composition, inferring functional diversity through FAPROTAX. Regarding pedological conditions, the three vegetation types exhibited significant organic carbon and pH differences. These distinctions were further validated by different microbial community compositions and functional potentials related to denitrification and nitrification, though not to nitrogen fixation. These findings reveal the interconnectedness between vegetation and soil microbial communities, emphasizing substantial implications for nutrient cycles. The gathered information holds practical value to define sustainable management of future grazing plan, considering agroecological principles.

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Reciprocal help, river flooding and a "Flying Herd". A project for the rehabilitation of Mongolian rangeland in a changing climate

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In this contribution, we present the preliminary results of an in-depth ethnographic research on post-socialist Mongolian herding practices. We aim to illustrate the value of an interdisciplinary approach merging cultural anthropology with animal production science in understanding the tactics herders employ in response to the evolving socio-economic and ecological landscape they inhabit. Currently, our research involves ongoing content, narrative, and discourse analysis of qualitative data collected in 2023 during seven months of participant observation and semi-structured interviews. This data has been produced with herders participating in a biodiversity restoration project within a degraded area in Bayangol sum, Selenge aimag. After categorising the data, we investigated language, communication, and day-to-day herding practices that herders enact among themselves, their animals, and the rest of the landscape, to analyse the formation of Pasture User Groups, that emerged in recent years all over Mongolia to counteract the atomisation of post-socialist pastoralism. While certain aspects of contemporary practices echo pre-socialist pastoralism, they are now shaped by the fusion of traditional knowledge, the prevailing socio-political climate, and exposure to financial and global markets. Additionally, we observed the limited influence of the shepherds' perception of the environment on the formulation of such projects, which has implications for their acceptance within the community. The flood of the river Kharaa provided an opportunity to investigate the herders' perception of climate change: a widespread awareness of the phenomenon did not translate into its linkage with the copious rain that brought the flood, whose causes were found elsewhere.

Roles, difficulties and needs of advisors in Climate-Smart Farming Transformations. Results from focus groups in 10 different European countries

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The European farming sector is facing climate change and is expected to reduce its Green House Gases emissions, store carbon in soils and landscape while adapting to a more extreme climate. In these on-going transformations, farm advisors are recognized as key actors in fostering climate-smart innovations and supporting farmers in transitions. The project ClimateSmartAdvisors is a pan-European multi-actor network started in 2023. Its aim is to boost the EU agricultural advisory community in the field of climate smart farming by strengthening the advisors' capacity and role in the transition towards Climate-Smart Farming (CSF). Focus groups with field's advisors were conducted in 10 different European countries in November / December 2023. The goal was to understand the roles, challenges, tools and advisory approaches of advisors taking into account the specificity of the different contexts. Despite differences in contexts and AKIS structures, advisors expressed similar views on their role in CSF transitions. Key aspects of their job emerged from the discussion such as the importance of building up a good farmer/advisor relationship, of developing an understanding of the globality of the farm, of involving the farmers in finding relevant measures for the farm and of supporting the farmers in implementing change. On these different aspects, advisors shared good practices, useful tools and advisory approaches but also their difficulties and needs in order to improve their advice. The needs expressed call for action at the level of the AKIS (develop interdisciplinarity, coordinate advice) as well as structural (economical compensation of environmental beneficial actions) and technical levels (develop hands-on description of beneficial CSF practices). This project has received funding from the Horizon Europe research and innovation programme under Grant Agreement No 101084179.

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Theatre 13

On-farm measurements of enteric methane emissions: Relationships between cow traits and emission intensity

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This research was carried out in a commercial dairy farm and aimed at measuring enteric CH₄ emissions and investigating relationships between selected animal traits and emission intensity. CH₄ emissions were quantified using a "sniffer" system (MooLogger, Tecnosens) installed in the automatic milking system (AMS) where an infrared gas analyser sampled the cow's breath through a flexible tube inserted into the AMS feeder. Sniffer data were measured in a continuum at one-second intervals from 64 lactating Holstein cows over one winter month. During the sampling period, the feeding management of cows was not altered. Effects of parity, milk yield, days in milk (DIM), rumination time and somatic cell count (SCC) on emission intensity per cow, calculated as daily CH₄ per kg of FPCM (ppm/kg/day), were evaluated. Preliminary results showed that the emission intensity was significantly affected by parity, milk yield, DIM and rumen activity. The analysis of parity showed a lower emission intensity for pluriparous compared to primiparous cows. Cows producing more milk emitted more CH₄, but emission intensity was significantly lower compared to low-producing cows. The lactation phase was also a significant factor affecting emission intensity. When considering the rumination time, the emission intensity was lower in cows with a higher rumen activity. Finally, SCC pointed out a non-significant positive trend, in that cows with low SCC were associated with lower emission intensity. Further data collection is ongoing to confirm these preliminary results and to validate the sniffer as a tool for monitoring enteric methane emissions in dairy cows.

Global prediction of heat stress-related risk in dairy cattle

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This study aimed at predicting the heat stress-related risk in dairy cows at global level. Critical Temperature Humidity Index (THI) thresholds of heat stress (HS) for milk yield were established through a literature review. The thresholds were mediated by latitude, meaning that at lower latitudes cows suffer HS at a higher THI, whereas at higher latitudes HS triggers at a lower THI. The thresholds were: 74 for equatorial-tropical areas (LAT. 0–30°); 68 for sub-tropical areas (LAT. 30–45°); and 65 for temperate areas (LAT. 45–60°). Climatic data at a high resolution of 0.25-degree grids were retrieved for the period 1985–2100. The daily average THI of the 30-year period 1985–2014 (CliNO) and up to 2100 at 25-year intervals for the projected Shared Socioeconomic Pathways (SSPs) climate change scenarios as defined by IPCC was calculated. The scenarios were: SSPs 2.6 where CO₂ emissions are cut to net zero around 2075; SSPs 4.5 where CO₂ emissions are falling but not reaching net zero by 2100; SSPs 7.5 and 8.5 where CO₂ emissions double and triple by 2075, respectively. The number of days in the year for the scenarios SSPs with a THI over critical thresholds (risky days) were identified and compared against the risky days observed in the CliNO; the differences were expressed as percentages. The number of days at risk of HS will rise globally. In the two less sustainable scenarios, this trend is expected to become more pronounced towards the end of the century, with increases in equatorial and tropical regions reaching 100%, whereas in sub-tropical and temperate regions, the predicted additional risky days will range from 20 to 30%. These data demonstrated the necessity of taking adaptation measures to the effects of global warming to guarantee animal welfare and preserve the world's milk supply.

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Poster 15

The Cyprus Bovine Breed: zootechnical characteristics of a valuable indigenous cattle breed

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This survey explores the indigenous Cyprus Bovine Breed, gathering data through questionnaires and empirical animal measurements of the Cyprus Bovine Breed Association. Eighteen farmers, holding 43% of the total animal population, participated in the survey and key aspects, such as farm demographics, housing, reproduction, welfare and feeding practices, were recorded. With a total population of 1202 animals (629 adult cows and 79 bulls), the typical farm has 35 (\pm 27) animals, with 1 or 2 designated breeding males. None of the interviewed farmers reported regular use of drug substances (i.e. antiparasitic or antibiotics) and all provides limited winter housing in primitive stables, showcasing robust health and resilience of this breed. The cattle primarily graze on natural pastures, with minimal concentrate supplementation, few weeks before slaughtering. Indigenous calves exhibit a natural suckling period of approximately 5 months. The age at first calving for cows is 29 months, with an average weight of 437 kg for heifers and 517 kg for adult cows. Calving intervals are observed at 13,2 months, with gestation length being 289 days, showing minimal complications or dystocia. Newborn calves weigh 31 kg (male) and 30 kg (female) and by the age of 6 months, male and female calves reach weights approximately of 162 kg and 149 kg, respectively, doubling in weight by 12 months (342 kg for males and 307 kg for females). With an average daily gain of 0,99 kg and usual slaughtering age between 18 to 22 months, weighing approximately 400 to 500 kg. The unique Cyprus Bovine Breed yield meat with distinctive qualitative characteristics and increased added value.

Evaluating the potential of methane reduction effects of different extracts from Chestnut and Quebracho in *in vitro* ruminal fermentation

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The livestock sector is a significant contributor to anthropogenic greenhouse gas (GHG) emissions, with methane (CH₄) emissions from ruminants being major contributors due to enteric fermentation. Tannins have emerged as a potential avenue for reducing enteric CH₄ emissions. This study aimed to evaluate the *in vitro* CH₄ inhibition potential of twelve different extracts from chestnut (C1-6) and quebracho (Q1-6). The experiment consisted of triplicate 24-h rumen fluid incubation with treatments (each at 1500 mg/L of total volume) directly added to the total mix ration (TMR) used as a fermentable substrate. The control group (CTR) consisted of TMR only. CH₄ production was measured using Gas Endeavour® following the modified technique by Tilley and Terry (1963). Results indicated a significant reduction in CH₄ production exerted by C1 extract (189 ml±24), decreasing by 20% compared to CTR (237 ml±55) (P-value <0.0001). C2 exhibited the most substantial methane reduction at 23%, while among the quebracho extracts Q6 showed a reduction of -22% (P-value 0.022 and 0.031, respectively). C3, C4, C5, C6, and Q1, Q2, Q3, Q4, Q5 did not show significant CH₄ reduction (P-value >0.05). Combining the most effective C extracts (C1+C2) yielded a significant methane reduction of -20%, with no cumulative effect; therefore, based on this result, a synergistic effect of extracts was not observed. Overall, results from this study suggest that C might be more effective than Q in reducing *in vitro* CH₄ production. Further research is warranted to fully understand the differential effects of various tannin extracts on CH₄ production in animal feeding.

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Poster 17

Animal genetic resources in Western Africa to support the agroecological transformation towards sustainable peri-urban farming systems

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Western Africa must navigate the challenge of ensuring food security, sustainable agriculture, and community well-being amidst urbanization and environmental deterioration. Peri-urban farming provides sustenance to urban areas, alleviates urban encroachment, and supports farmers' livelihoods. In communities where farming is integral, adopting One Health approaches is crucial to address emerging public health issues. Agroecology emphasizes ecological principles, biodiversity, soil and environmental health, and climate change resilience. Utilizing local animal genetic resources is imperative for agricultural sustainability in the region, as indigenous breeds exhibit unique resilience traits. In this context, a survey was conducted to identify local genetic resources and traits of interest. The local breeds identified were the dual-purpose Goliath and Fulani ecotype chickens, the Ashanti Black Pig, the West African Dwarf and Sahelian goats, the Sahelian sheep and the Gobra cattle. Key traits of these breeds include resistance to diseases, adaptation to water and food scarcity, and tolerance to heat stress, humidity and aridity. Integrating peri-urban farming, agroecology, One Health, and preserving local genetic resources is imperative for sustainable agriculture and food security. This interconnected approach not only addresses current challenges but also sets the foundation for resilient and adaptive farming systems that can thrive in the dynamic urban and environmental landscape of the region. Funded by EU under the "URBANE – One Health approaches to support agroecological transformation of peri-urban farming" project (Grant No: 101059232).

Multifunctionality of sheep grazing in marginal areas: a remote sensing approach in Eastern Italian Alps
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Mountain pastures deliver both provisioning and non-provisioning ecosystem services. However, incorrect management may diminish their capacity to provide these positive externalities. In a pilot project aimed at restoring an abandoned alpine pasture and valorizing its biodiversity and cultural ecosystem services, this study utilized remote sensing and GPS tracking to assess the impact of sheep grazing on the pasture's primary production. The study was conducted in the summer of 2023 in a pasture that had remained ungrazed for three years at 2000 m asl (Monte Coppolo – Eastern Italian Alps). The pasture was divided into four one-hectare sections, grazed by a flock of 30 “Lamon” local breed sheep using a rotational system with 2-week periods. GPS collars on three sheep monitored spatial use intensity, with positions recorded every two minutes. Specific spectral indexes obtained from the Copernicus SENTINEL-2 satellite mission were used to estimate the primary productivity with a pixel size of 10 m². We used NDVI (Normalized Difference Vegetation Index), GWI (Green Way Index) and Delta NDVI (Maximum NDVI peak of 2023 – NDVI September 30, 2023). Animals' activity was higher during daytime compared to night-time. The spatial intensity of use of the plots was uneven and showed a preference for areas with gentler slope. The combined analysis of spectral indexes and spatial intensity of use revealed vegetation regrowth patterns in intensively used areas at the end of the growing season. The results of this first year confirm that combining remote sensing and GPS tracking for monitoring alpine pastures could represent a valid contribution to analyzing the positive outcomes derived from these agroecosystems.

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Poster 19

Genome-wide scan reveals candidate genes related to milk production and adaptive traits in Sicilian cattle breeds

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Selection towards heat-tolerant traits represents one of the main challenges of the livestock sector. Preserving the diversity of local breeds such as Cinisara (CIN) and Modicana (MOD) cattle, native to Sicily and adapted to hot climate, can be crucial for enhancing the resilience of the species. Using the BovineHD BeadChip, we genotyped 64 animals (26 CIN, 38 MOD) to assess within-breed genetic diversity, runs of homozygosity (ROH) patterns and pairwise FST-outliers to identify potential genomic regions associated to resilience and heat tolerance. Our findings indicated a moderate level of genetic variability within both breeds. CIN exhibited half of the genomic inbreeding of MOD (FROH = 0.06 vs 0.12). The prevalence of short ROHs highlighted the occurrence of old inbreeding events. Seven ROH islands in CIN harboured 643 markers distributed in 7 chromosomes, whereas a unique ROH island on BTA6 (783 SNPs) was identified in MOD. ROH islands and FST-outliers approaches revealed QTLs involved in the milk production, reproduction, health, and local adaptation traits. In CIN the ROH hotspot on BTA14 (57 SNPs) harboured QTLs and genes (HSF1, DGAT1, ZNF34) related to milk production and heat tolerance. In particular, the HSF1 gene plays a key role in the cellular response to heat stress in lactating dairy cows. These initial findings encourage further study to disentangle the genomic basis of the adaptation capabilities to hot climate conditions in the Sicilian native breeds.

National Livestock Biobank: an initiative supporting the valorisation of the Italian Livestock genetic diversity
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Genetic and genomic scientific advancements in livestock rely on the availability of biological specimens. The LEO project (PSRN 16.2 Livestock Environment Open data) established a National Livestock Biobank (Biobanca Nazionale Zootecnica-BNZ) hosted by ConSDABI Research Center with the goal of creating a comprehensive ex-situ biological and germplasm bank along with associated data (e.g. animal id, breed, sex, age etc). BNZ aligns with the FAO's Global Plan of Action for Animal Genetic Resources, establishing ex-situ conservation programs and developing technical standards (Strategic Priorities 9-11). Equipped with advanced technology and modern infrastructure, BNZ preserves a wide range of biological samples from numerous species and breeds of farm animals. BNZ' current archive counts over 630,000 biological samples, with a flow of forty-thousand new samples per year. Livestock biodiversity in BNZ is represented by 79 dairy and beef cattle breeds, two buffalo breeds, 84 sheep and 60 goats breeds, 80 equine breeds, 25 swine breeds, and 46 rabbit breeds. Samples span more than 30 years. Currently, BNZ receives and manages about 232 sample requests per month. Indeed, BNZ provides a significant resource for scientific research, biodiversity conservation, and food security, and it should be supported and further developed to maximize its positive impact.

Session 52

Poster 21

Participatory workshops to design innovative mixed farming systems based on the synergy between livestock and viticulture in Burgundy

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The integration of crop and livestock systems is increasingly recognized for its potential to enhance ecosystem services. The synergy between livestock production and viticulture is described as a promising example of an agro-ecological integrated crop–livestock system. However, this integration is challenging due to technical and organizational issues requiring collective re-design. Two participatory workshops were carried out involving breeders, wine growers, winegrowers who use animals in their vineyards, viticulture and livestock stakeholders, and researchers in Burgundy interested in the ecosystem services provided by this synergy. In the first workshop, participants discussed the forms, benefits, and constraints of this integration, addressing feasibility concerns. In the second workshop, expert support was provided, and two sets of experimental designs prototypes were designed to address technical aspects. Participants' thoughts were organized into two types of ideas: technical ideas focusing on vineyard treatments' impact on animal health, crop cover types, input use, and organic matter supply and organizational ideas. Thus, participants highlighted the importance of territory development, animal surveillance, exchange organization between wine growers and breeders, and contractual agreements. The prototypes designed in the second workshop aimed to address a common question defined and validated by all the participants regarding crop cover types and management strategies to preserve vineyard and animal health while meeting animals' nutritional requirements. The first group proposed testing various temporary crop covers, while the second suggested testing a permanent cover. Both groups recommended evaluating the prototypes using a multicriteria assessment method. Further collective redesign has to be organized to develop a virtual platform to facilitate communication between breeders and wine growers.

Landscape characterization of Italian small ruminant populations

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Livestock farming is strongly interconnected with the surrounding environment, especially for small ruminants, which are often bred in extensive systems. Thus, characterizing the landscape of these farms offers insights into their ecological impact and conservation implications. Public data and maps about Italian ecosystems, land cover, soil use, landscape types, and naturalistic-cultural value were retrieved from ISPRA. We geolocated 3851 registered farms, raising 41 goat and 47 sheep Italian populations, and assessed the composition of their surrounding areas within a 10km buffer. At a species level, “Agricultural areas” -most represented in Southern breeds- and “Forest and woodland” -most represented in Northern breeds- accounted for about 75% of the ecosystems occupied by both sheep and goat farms. Among land covers and soil use, “Broad-leaved trees” and “Forestry use” were prevalent in both sheep (38%) and goats (43%), but Southern sheep breeds also covered a relevant part of “Arable lands”. Goat farms were predominantly in mountainous areas, while sheep farms spanned mountainous (38%, especially in the North) and hilly (31%, especially in the South) terrain. Lastly, sheep farms were situated in areas with an overall lower naturalistic-cultural value than goats. This study delineates the distribution of Italian small ruminant farms and the characteristics of the land they occupy, highlighting differences among species and breeds. These data might help to develop management strategies to valorize both Italian animal and environmental resources and facilitate the harmonious coexistence of livestock farming, ecosystem health, and biodiversity preservation.

Session 52

Poster 23

Evolution, typology and issues of pig farms in France: lessons from the 2020 agricultural census

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Every 10 years, the agricultural census provides an overview of French agriculture by collecting more than 900 datapoints from all agricultural holdings (AH). In 2020, mainland France has 389,779 AH (-20% in 10 years). Half have a livestock activity, which is significant in 167,000 AH (-24% in 10 years). Of the latter, 82% rear herbivores without significant granivore production, 10% rear granivores without significant herbivore production, and 8% are mixed. The decrease in the number of AH with at least one pig is slowing down: 59,549 in 2000, 22,286 in 2010 (-63%), and 13,048 in 2020 (-42%). Nearly all pigs are reared on 8,448 AH that contain more than 100 pigs or 20 sows, of which 4,426 have sows. This number was twice as large in 2000. Farrow-to-finish farms remain largely dominant in France, with 80% of sows and two thirds of fattening pigs of the country. The decrease in the number of pigs is also slowing down: 13.3 million pigs in 2020 (-3.5% from 2020-2010 vs. -7.1% from 2010-2000), including 955,795 sows (-14.3% vs. -21.3%). With 56.3% of France’s pigs, Brittany remains the leading pig-production region, but the decline in the number of pigs is greater there. Three main types of pig farms coexist in France: (1) Specialized (44% of farms and 67% of pigs in 2020, 39% and 61% in 2010), (2) Crops and pigs (20% of farms and 11% of pigs, unchanged), and (3) Herbivores and pigs (29% of farms and 21% of pigs, 38% and 27% in 2010). The evolution towards production increasingly specialized in pigs, with little or no agricultural area and without producing other types of animals, even sows, reflects the difficulty in reconciling several productive activities but also, more certainly, separating the activities of agricultural companies that have specific legal structures.

Two methods for detecting roe deer fawns before and during mowing, resp.

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Young roe deer fawns can be injured or killed during mowing of grassland and fodder crops in May and June. The aim of the study was to evaluate the achievements of two methods for detecting fawns before and during mowing, resp.: the portable device with infrared sensors called “Wildretter” manufactured by isa industrieelektronik GmbH (WR; carried by the operator or fixed on a quad bike), and the assistance system SENSOSAFE manufactured by PÖTTINGER Landtechnik GmbH (ASS; mounted directly on the front mower and the front hydraulic system for the rear mower), which utilises the interaction between optical sensors and an integrated light source. Data, such as correct fawn detection and failed fawn detection (i.e. the number of fawns seen or found during or after mowing and the number of fawns found injured or killed after mowing using WR and ASS, resp.), were collected at 13 and 9 days, and from 21 and 36 plots using WR and ASS, resp. A total of 10 (9 detected by a person) and 6 fawns (1 detected by a person) were located at 5 and 3 plots by searching with WR and ASS, resp. Unfortunately, the mower did not stop in time once when a fawn was detected by sensors using ASS. Moreover, 2 fawns (one injured and one killed) were found after mowing with ASS, one of them undetected due to a missing signal connection after new ASS installation. No fawn was seen or found on the plots after searching with WR. In conclusion, not all fawns could be detected using ASS. However, due to the utilisation of infrared sensors by the WR device, the reasonable performance of this device could be observed only during early morning hours, whereas ASS can be applied around the clock directly by mowing.

A survey on young post-graduated farmers: profiles, motivations, and difficulties

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The French agricultural sector is currently facing a major challenge in terms of renewal of generations. With 25% of French farmers set to retire before 2030, the setting-up of new farmers is crucial. The present study focused on farmers who obtained a master’s degree and started their activity for less than 10 years. Forty-one of them were identified by alumni network and surveyed by master 2 students of the Institut Agro Rennes-Angers. Qualitative semi-structured interviews were performed using a pre-established interview guide. Four profiles (groups A, B, C and D) of farmers were established based on their motivations for setting up, using multiple correspondence analysis (MCA). Group A (n = X) was motivated by the desire to play an active role in promoting healthy production, mostly with on-farm processing and direct-sales. Group A included breeders set up outside the family, choosing local breeds. Group B were motivated to maintain the family farm (n = X). They were not involved in certified production; they reported limited holiday time (< 7 days/year) and use more outsourcing than other profiles. Group C (n = X) expressed optimism about the future of farming. They set up independently, they employed staff to manage farm operations and were partially satisfied with their income. Group D (n = X) primarily sought challenges in their live and were mainly specialised in monogastric species. Overall, results from this study may provide insights to facilitate future setting-ups from these profiles. A positive aspect in this study is that these master’s graduate farmers demonstrated a strong willingness to exchange and share their experience.

Dechoriation of Honey Bee Embryos by Alkaline Solutions

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Honey bees are strategically important to nature since they are primary pollinators and major contributors to economy. Sadly, populations of honey bees are declining by agricultural spraying, electromagnetic radiations, bee diseases, etc. Therefore, it is important to find a preservation method for honey bee gametes and embryos in order to challenge to the colony losses. Due to the chorion membrane of the eggs which control water loss by regulating the water exchange, it extremely challenging to transfer any form of agent that is necessary for the successful preservation into the eggs. The effects of treating honey bee eggs with alkaline solutions that is used for dechorio-iation were assessed in this study. With some modification, the dechorio-iation process is conducted according to Li et al. (2010). Untreated eggs (C) were compared with alkaline solutions (%1,5 Sodium Hypochlorite (SHC), %1,5 Sodium Hydroxide (SHD) and Hexane (HX)) treated eggs for the development. A total of 327 honey bee eggs (0-18 h age) is used to evaluate the effects of alkaline solutions on the further development to larva stages. The results showed that the eggs continued their development similar to control group (C, 93%) and progressed to the larva stages (SHC, 78% and Sprey-HX, 85%; $P>0,05$) by using 1.5% SHC and the Sprey-HX method. Furthermore, the application of 1.5% SHD resulted in a 40% rate of alive larval development. Additionally, the study showed that direct application of Hexane onto the eggs via pipette caused a complete hold in egg development. As conclusion, this study demonstrates that treatment of honey bee eggs/embryos with spray-hexane, sodium hypochlorite, and sodium hydroxide allows for their continuing development.

Session 53

Theatre 2

The effect of a regional legislative act on the distribution of honey bee mitochondrial DNA lineages

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A few national or regional legislative acts or initiatives have been introduced in Europe to preserve the genetic integrity of native honey bee subspecies. In Italy, for example, the region Emilia-Romagna (North of Italy) has been one of the first regional authorities that issued a regional law, specifically addressed for this purpose, focused on the conservation of the genetic integrity of *Apis mellifera ligustica*, banning the breeding and introduction in the region of other subspecies. The relevance of this regional law is derived by the fact that this region hosts one of the highest concentrations in the world of the queen breeding activities for *A. m. ligustica*. In this study, we monitored the distribution of mitochondrial DNA (mtDNA) lineages in a total of 1143 honey bees, each collected from different colonies, taken from apiaries spread all over the region Emilia-Romagna and over three years (2020-2022), after the entry of the mentioned regional act. The informative mtDNA region spanning the tRNA^{Leu}-COII genes was sequenced for all these samples and a total of 14 different mtDNA haplotypes (mitotypes) were identified. The most frequent haplotype (86.5%) was C1, which is characteristic of *A. m. ligustica*. The second mitotype considered to characterize this subspecies (M7) was identified in very few samples (<1%). This study indicated that the regional act seems to be effective in strengthening the conservation of *A. m. ligustica* genetic pool. Acknowledgements: Funded by regione Emilia-Romagna, BEE-RER projects.

A Genomic Approach for the Creation Of Isolated Mating Areas for the Conservation of Italian Ligustica and Carnica Honey Bees

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Beekeeping is dramatically facing the effects of globalization. The results can be seen in the progressive diffusion of non-native varieties of *Apis Mellifera* (A.m) in areas where these were not originally present. The reproductive mode of bees (in flight) poses a serious problem of conservation and selection of local geographical varieties. Therefore, the introduction of mating control in beekeeping can be seen as a suitable conservation approach to preserve biodiversity. The aim of this study was to create three Isolated Mating Areas (IMA) for the Italian honeybee's population of Ligustica and Carnica bees, using next-generation sequencing (NGS) techniques for breed assignment. The project (SESALMEC) involved the identification of specific isolated areas such as islands or valleys. Subsequently, numerous beekeepers submitted their best mother colonies to a test of conformity to the genetic type of interest. In total more than 60 colonies were tested based on whole genome sequence analysis. Thirdly, several sister Virgins Queens (Q) were produced by each of the mother colonies (M) that passed the test. These colonies were then located in the isolated mating areas and committed to the production of drones. The results of the project are the creation of two isolated areas for the Ligustica bees in San Pietro Island (South West Sardinia), Ponza Island (LT) and a third isolated area for the Carnica bees in the Val Bodengo valley (SO) in the north of Italy. The conservation areas are active since 2022 and protected from the presence of other colonies of unknown variety by specific local ordinances. Interested beekeepers can bring their own virgin queens for mating.

Session 53

Theatre 4

Using genomic information to monitor diversity of the Australian honeybee genetic resources

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Modern animal breeding technologies combining statistics and genomics can provide tools for generating reliable genetic improvement in a wide range of traits but have not yet been applied to honey bees in Australia. Genetic improvement of honey bees will allow the continual identification and use of queens that will permanently and continuously improve traits that are important to the honey bee industry (such as pest and disease resistance, honey production, pollination performance, and temperament). Using genomic data within the context of a honey bee breeding program can identify genetic relationships between individuals to increase accuracy of breeding values. Genomic data can also provide information about managed honey bee populations, to estimate stratification and introgression levels and to understand complex population admixture events or identify signatures of natural and artificial selection. For the majority of livestock species genomic data are obtained through the use of commercially available Single Nucleotide Polymorphism (SNP) array chips, which can provide accurate and evenly distributed SNPs with known locations across the genome. Such chips have only recently become commercial for bees. SNP genotyping can be easily reproducible across different batches of samples and is tolerant of lower quality DNA extracts. In this study, we investigate using DNA extracted from pooled drone samples and imputed genotypes from low pass sequencing for genomic prediction and population structure analysis.

Exploring shifts in the microbial community when rearing different genetic lines of black soldier fly in a single production facility.

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The potential of farmed edible insects to improve our food system's circularity is what is driving a fast-growing sector. At the same time, significant gains can, and need to, still be made with respect to insect production efficiency to maximize their circular potential. To this end, selective breeding has already proven to be effective to boost production in plant and livestock. Its potential is however only now being explored and exploited in the field of farmed insects, like the black soldier fly (BSF). Yet, key fundamental questions, important to the implementation of breeding programs remain. One such question is whether the resident microbiota of a specific insect strain should be taken into account when comparing different genotypes. To assess this, the microbiome of ten BSF strains was mapped using 16S rRNA gene sequencing for the adult stage at two time points, (i) when the strains first entered our rearing facility and (ii) after a number (between 6 and 11) of consecutive rearing cycles. The larval stage following the second adult sample point was also analyzed. While most zOTUs (zero-radius Operational Taxonomic Unit, representing a unique DNA sequence and thus member of the microbial community) were found to map to Firmicutes and Proteobacteria for the adult at both sampling points, the ratio between both phyla, as well as the present genera of these phyla, varies in-between strains. For example, the unidentified Enterobacterales (zOTU11) is extremely abundant (almost 40% of the reads) uniquely in one strain at the initial time point. Even after multiple rearing cycles, the microbial communities remain distinctive for each of the strains. At the larval stage the actinobacteria phylum was also frequently present, aside from the two phyla dominating the adult stage. Our findings reveal that the microbial community does not fully converge between strains, even when reared in identical conditions, warranting care when interpreting phenotypic output during selective breeding programs.

Session 53

Theatre 6

Consecutive mating of black soldier fly males to produce full- and half sibling offspring

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In animal breeding programs, utilizing quantitative genetic designs such as full/half sibling design is fundamental. This however demands that mating can be controlled, including having individual males mating with several females. In the literature, black soldier fly (BSF) males and females are reported to have to gather in lekking groups to engage in competitive displays and courtship rituals before they will mate. This lekking behavior is described as crucial for establishing suitable mating conditions, and mating of BSF pairs or individual sires is therefore assumed impossible. We show that an individual virgin male can be successfully mated with an individual virgin female. Furthermore, we show that an individual male can mate several virgin females, in our experiment up to four, consecutively within up to four hours and produce eggs and offspring with each mated female. Our findings pave the way for moving beyond mass selection to now include pedigree information in BSF breeding. This has the potential to allow selection for multiple traits simultaneously, control inbreeding, and increase rates of selection responses compared to phenotypically based mass selection.

A bio-economic model for estimating economic values of important production traits in the black soldier fly (*Hermetia illucens*)

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Insects are a promising source of sustainable high-quality protein for feed and food. The production of insects could be optimized by using selective breeding tools, such as a breeding goal to select the best parents. A breeding goal is a collection of traits, where each trait is weighted with a so-called economic value (EV). This study is the first to investigate the EV of important production traits in an insect species, namely, the black soldier fly (BSF). We used a bio-economic model to estimate EV for larval mass on day 15, feed intake, growth rate, dry mass-, protein- and fat content, development time, eggs per fly, egg hatching rate, and larval mortality. The EV of a trait was the increase in profit when the trait value was increased by 10%, while the other traits were unchanged. The results revealed that the traits with the highest EV were those related to composition (dry mass and protein). The lowest EV were for traits related to reproduction and development time. Before we can implement a breeding goal in a BSF breeding program, we need to learn more about the genetic parameters of the most relevant production traits. In conclusion, the EV presented in this study are the first step in the development of a more sophisticated insect breeding program.

FLYgene: Advancing Sustainable Breeding Programs and Genomic Tools for Black Soldier Fly (*Hermetia illucens*) in Kenya and Uganda

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The FLYgene project is aimed at advancing our knowledge of Black Soldier Fly (BSF) genetics, genomics, and phenomics, with a primary objective of implementing sustainable breeding programs in Kenya and Uganda. This project is designed to achieve multifaceted goals, including identification and prioritization of BSF traits in smallholder and commercial production systems, development of large-scale phenotyping systems, and design breeding programs tailored to production systems. FLYgene develops genomic tools for marker-based monitoring of genetic diversity and pedigree tracing, facilitating tracking of BSF lineage. FLYgene has achieved notable milestones, conducting extensive surveys to characterize BSF production systems and trait preferences in Kenya and Uganda. Computer-vision-based approaches have been prototyped for precise phenotyping of BSF larval traits. Whole genome sequence data from ~300 BSF, sourced from wild and captive environments, provide insights into the genetic diversity. Protocols have been developed for the pilot implementation of mass selection at private-sector entities to drive innovation in BSF breeding practices. FLYgene, funded by the Ministry of Foreign Affairs (Danida), Denmark, is characterized by a strategic and impactful collaboration between multidisciplinary researcher teams and private-sector partners. Encompassing five PhDs, FLYgene is ensuring long-term capacity building.

Exploring genetic architecture of larval body weight in Black Soldier Fly

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The knowledge on genetic architecture of larval body weight in Black Soldier Fly (BSF, *Hermetia illucens*) is limited. The genetic improvement program established by Protix and Hendrix Genetics resulted in a remarkable 39% increase in larval body weight within the initial two years. It has now been running for over 40 generations, with biobanked flies for each generation. Our aim was to detect regions of the genome and potential genes affected by genetic selection. DNA was extracted from 30 to 90 flies every four generations from base population to generation 28. Samples were pooled within generation and sequenced with 30X coverage. Following quality control, variant calling and filtering, 18 million SNPs (single nucleotide polymorphism) were identified. The SNP frequencies were determined utilizing the PoPoolation2 toolkit. Wright's Fixation index was computed for all SNPs across pairwise combinations of generations. Chromosome regions were identified applying a meta-analysis approach that integrated multiple methodologies, including average FST calculations on 10kb windows, smoothing spline techniques according to the evolution of FST on neighboring SNP, and average FST by gene for the known genes in NCBI database. This analysis pinpointed 170 genes within nine regions across six chromosomes. Further analysis was conducted through an enrichment gene analysis using Shiny GO, which revealed a significant enrichment of genes associated with developmental and primary metabolic processes. These findings shed light on the genetic mechanisms governing larval body weight in BSF and offer valuable insights with potential implication for enhancing genetic improvement efforts in this economically important species.

Session 53

Theatre 10

Genetic parameters for house fly larval performance in two dietary environments

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Optimal breeding of insects requires genetic parameters for production traits. Performance may vary with the production environments, emphasising the need to investigate variance components and their interactions with environmental factors such as dietary regimes. In this study, a full-sib half-sib mating design was employed to estimate variance components for individually recorded larval size, dry mass, and fat content in an outbred population of house flies (*Musca domestica*) as well as correlations among these. 200 paternal half-sib groups with two full-sib groups each were established. The full-sib groups were reared either on a standard laboratory medium or a diet consisting of grass-protein waste. Variance components and correlations were estimated using uni- and bivariate linear mixed models for the traits measured within and across the dietary environments. We report variance components for all recorded traits as well as correlations between traits and unveil large effects of fixed and random non-genetic variation on total phenotypic variation, providing useful information for the design of optimal breeding schemes.

The effect of adult's sex ratio on the reproductive output of *Tenebrio molitor*

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Manipulating sex ratio has been studied thoroughly as a means of more efficient mass rearing of insects. Recently, the approach of mass production is also applied in the insects as food and feed. Although much research is currently directed to the optimization of larval growth parameters, a factor that has not been thoroughly investigated yet is the effect of sex ratio on the fecundity of farmed insects. Findings so far concerning other insect species have shown that the optimum sex ratio is species-dependent. Based on that, in the present study the effects of different percentages of female:male ratios (50:50, 60:40, 40:60, 80:20, and 20:80) and adult densities (10, 20 and 30 adults/vial) on the reproductive output of the yellow mealworm, *Tenebrio molitor*, were evaluated. Adults were left undisturbed to mate and oviposit for 7 d. After this interval, adult mortality was recorded for both males and females, while laid eggs were collected and counted. After an additional interval of 14 d, the number of the newly-emerged larvae was recorded. The same procedure was repeated five times for a time period of 28 d in total. The results showed that both factors affected offspring production. Interestingly, for density 20, 20% females produced more offsprings per female. These results tend to reveal the optimum potential of the population, promoting the mass production of *T. molitor*.

Session 53

Theatre 12

Response of selected full-sib yellow mealworms to diet and density modifications : a GxE analysis

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YnFABRE project aims to develop a breeding program tailored for *Tenebrio molitor*, from the creation of genetic gain in breeding nucleus to the delivery of new genetic lines into productive vertical farms. Environmental conditions in the breeding nucleus, which optimize performances at family level and at low-density scale, differ from those in farms, emphasizing the mass rearing of mealworm populations. The risk is therefore that pedigree-based family selection might be ineffective under the conditions on the farm. Knowledge are so required to estimate a potential genotype – environment interaction (GxE). Full-sibling larvae from 20 families chosen according to their EBV for growth, have been reared under various density and diet conditions (0.1, 0.4, 0.7 g of larvae/cm² under high and low energetic diets). All larvae were monitored during their exponential growth phase and average biomass daily gain was recorded. GxE analysis will be conducted by comparing family performances and ranking across different conditions. Preliminary results highlight a density effect on the larval viability (increase of 35% of dead larvae for 0.4 and 0.7g/cm² compared to 0.1g/cm²), without any significant difference on larval growth. The experiment is in progress and based on the full results, the environmental conditions of the nucleus could be adjusted to mimic the farm's one and limiting the GxE effect.

Genetic parameters estimation in yellow mealworms: A comparison of pedigree and genomic approaches
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The study aimed to compare the genetic parameters estimates from pedigree and genomic data in yellow mealworms. We analyzed the performance of 15,000 mealworms divided into 975 sib-groups, across 3 generations, for 20 traits (reproduction, growth-feed efficiency, survival at the sib-group level; and individual weight and developmental time). Among these, 3,451 individuals were genotyped using the high-density Axiom YNS_Moll array, genotyping 675K SNPs. Genetic parameters were estimated using linear animal model based either on pedigree data exclusively, or a combination of pedigree and genomic information. Some variations in genetic parameters estimations were observed depending of the employed approach- (pedigree or genomic), and standard errors seem lower under genomic evaluation. For instance, in the case of the developmental time between egg to pupa, the heritability estimate was equal to 0.18±0.04 when utilizing pedigree information alone, while it increases to 0.30±0.02 when integrating genomic data. Furthermore, the heritability estimates for sib-group phenotypes seem to be overestimated with only pedigree data. In conclusion, despite the cost of genomic evaluations, the use of genotype information promises in enhancing the accuracy of selection scheme of some criteria for insect breeding programs.

Genome-wide association study in *Tenebrio molitor* reveals reproduction, development and growth quantitative trait loci

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Tenebrio molitor, also known as the yellow mealworm, is farmed as a new alternative and sustainable source of protein and is valued for its high nutritional content and ability to thrive under various conditions. Identifying genomic regions, genes and markers associated with agronomic traits will be a first step for genomic selection. Our study aims to understand the genetic basis of complex traits involved in growth, development and reproduction through genome-wide association studies (GWAS). We performed single and multi-locus GWAS methods on 136 individuals genotyped by whole-genome high-throughput sequencing and phenotyped for 26 quantitative traits involved in growth, development and reproduction. The analysis of more than 5 million of markers allowed the identification of several significant QTLs and their associated genes. The genomic regions identified and their associated molecular markers represent a key step towards breeding programs for the improvement of yellow mealworm production.

Comparing expected and observed relationships among a sample of Slovenian honey bees

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Recently, there has been an increased interest in genetically improving honeybees through systematic breeding programmes. Active monitoring of the population genetic diversity is essential to manage population diversity and counteract the adverse impacts of inbreeding. This work aimed to compare pedigree (expected) and genomic (observed) relationships among colonies using SNP data. We collected 288 Slovenian *Apis mellifera carnica* honeybees in summer 2023, from the isolated Alpine valley (Krma), as part of the BeeConSel project. Four drone producing colonies (DPCs), presumed to be the only males present in the isolated area, were installed prior to 9 virgin colonies being brought for mating. We genotyped all queens, 30 workers per queen and a pooled sample of the 4 DPCs on a SNP chip 4165 SNPs. After quality control, we had genotypic data for 8 queens, 235 workers, and 4 DPC pools, each with 1781 SNPs suitable for analysis. We used multiple pedigree parentage assignment software – Sequoia, COLONY2, AlphaAssign, and R code based on opposing homozygotes – to reconstruct pedigrees from the SNP data whilst comparing the outcomes. This allowed us to identify which DPCs were most likely to have produced the workers' fathers and to inspect the relatedness within and between the colonies. Notably, the multiple pedigree software outputs indicate that some workers received no assigned father, suggesting that unknown colony drones were able to mate with the queens, underscoring potential isolated mating area errors. From the reconstructed pedigree and SNP data, we calculated pedigree and genomic relationship matrices to compare relatedness within and between colonies and differences between expected and observed relationships due to recombination, segregation, and background relatedness. Our work demonstrated various methodologies for analysing SNP data, evaluating their accuracies, running times and outputs. This research is valuable as the conscious monitoring of genetic diversity in breeding programmes is essential in promoting both long and short-term genetic improvements, ultimately advancing toward the desired objectives.

Genetic resources conservation programme of the Carniolan honeybee (*Apis mellifera carnica*) of the Dobra line in Poland

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The Carniolan honey bee (*Apis mellifera carnica*) is a subspecies of the Western European honey bee adapted to specific, often mountainous habitats. It is commonly known in Poland as Krainka and naturally occurs here. The Dobra line (*Apis mellifera carnica* L) is found in the Beskid Wyspowy range in areas covered with fir forests. Through natural selection, bees belonging to this line have adapted to local, harsh climatic and forage conditions as well as developed some adaptive traits, such as good winter hardiness, adaptation to overwintering on honeydew, limited egg laying in September, careful development in spring and rapid increase in numbers once the weather stabilizes. Thanks to the beekeepers' initiative, it was possible to establish a region for conservative breeding of this line in the Dobra area. The main program's assumption is based on creating an area for its conservation breeding in the form of an enclave with natural boundaries of mountain ranges. Thanks to this spatial isolation and the active protection provided by the beekeepers for decades, the local bees' phenotype has remained unchanged. Border-created administrative areas of conservation breeding for the Dobra bee include Dobra and Tymark communes. It is estimated that approximately 2,000 beehives are kept in the closed area. The Dobra line has been recognized and protected by a genetic resources conservation programme in Poland since 2014. In the year the program was launched, 1 leading apiary and 5 cooperating ones joined, maintaining 308 bee colonies. Currently (2022), the program covers 1 leading apiary and 7 cooperating ones, supporting 457 bee colonies. Hence, the population of Krainka of the Dobra line remains stable.

Morphological evolution of the female reproductive system of *Hermetia illucens* (Diptera: Stratiomyidae) after mating

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The Black Soldier Fly (BSF) offers a sustainable solution for converting organic waste into protein-rich animal feed. While its economic value is clear, knowledge gaps remain regarding its reproductive biology, impacting mass rearing efficiency. The objective of research was to investigate the morphological evolution of the female reproductive system after mating. Females were reared under controlled conditions and dissected throughout their lifecycle. The following morphometric traits were measured: Ovary Length (OL), Ovary Width (OW), Ovary Index (OI), Ovary Mass (OM), Egg Load (EL) and Adult Body Mass (ABM). EL was based upon a count of all mature eggs in each fly's reproductive tract. Also, we calculated the ratio of OM to ABM. All quantitative data were analyzed using the IBM SPSS Statistics software. After mating ovarian development in BSF goes through four stages. Comparisons of morphological traits between the different stages were described. Peak egg load and optimal discard age were estimated. Post-oviposition, ovaries appeared significantly smaller ($p < 0.001$) and yellowed. This study provides new insights into BSF reproductive biology, enabling the insect industry to optimize breeding processes and improve BSF production efficiency.

Small scale rearing for the development of genomic resources in black soldier fly (*Hermetia illucens*, Linnaeus, 1758)

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The black soldier fly (BSF) industry is growing rapidly. As with other livestock sectors, developing high-performance strains will allow for more effective production. Unfortunately, polyandry makes classical techniques of genetic selection nearly impossible to implement in BSF. Our project aims to develop a parental assignment tool based on the use of SNP-type genetic markers. The first step was to design a method of production that allowed control of the genealogy of each specimen for multiple production cycles. For each family at each generation ($n = 5$), 200 adults were maintained in complete darkness from emergence until mating. A total of 24 families were created at two experimental sites (university (A) and industry (B)). The first four mating couples of each family were isolated in their own containers. Collected egg clutches were suspended above Gainesville diet (ad libitum, 70% RH). At the neonate stage (5-day-old), only the larvae ($n = 200$) of one mating pair of each family were retained as the parental couple of the next generation. The phenotypic data of the first 4 generations (G) were analysed for trends between generations. Upward trends were identified for the pre-oviposition period at the B site (G1: 2.96 ± 0.80 days; G4: 3.44 ± 0.96 days) and the pre-hatching period at both sites (G1: 2.4 ± 0.8 days; G4: 3.1 ± 0.8 days). No overall trend was observed for the mean weight of 5- (A: 79.2 ± 25.1 mg; B: 53.9 ± 21.8 mg) and 10-day-old larvae (A: 235.0 ± 29.8 mg; B: 212.2 ± 26.9 mg) at either site. These preliminary results indicate that extreme inbreeding might influence the reproduction cycle without affecting the larval growth performance.

Impact of Low-Quality parent and interchanged diets on Black Soldier Fly (*Hermetia illucens*) Larval Traits and Microbiome Composition*A. Gligorescu¹, S. Nazipi Bushi², A. Schramm², J. Sørensen¹**¹ Aarhus University, Department of Biology, Section for Genetics, Ecology and Evolution, Munkegade 116, Building 1540, 8000 Aarhus C, Denmark, ² Aarhus University, Department of Biology, Section for Microbiology, Munkegade 116, Building 1540, 8000 Aarhus C, Denmark*

The production of insects on single sourced low-quality diet is of high interest since this can tackle critical waste management challenges and generate sustainable protein products. However, the use of low-quality diets have negative consequences on insect performance. The negative impact might be overcome over time by adaptive responses in the insect or the associated microbiome, however the mechanisms and consequences of diet adaptation are not well understood. This study investigates the impact on the performance and the microbial composition of black soldier fly larvae (*Hermetia illucens*) during a diet interchange experiment post diet adaptation. After 30 generations of being reared on either high-quality chicken feed (CF) or low-quality single source wheat bran (WB) diets, larvae underwent a diet interchange experiment for three generations. During this interchanged experiment, a total of six populations of BSF maintained on the parent diets (3 xCFp and 3x WBp) and six populations maintained on the interchanged diets (3x CFi and 3xWBi) were investigated. Trait measurements (e.g. larval weight, growth rate, pupa weight, female weight, fecundity and egg production) and 16S amplicon analysis was conducted on the 1st and 3rd generations of the interchanged experiment on different life stages (Eggs, juveniles, harvest larvae). Our results reveal differences across dietary treatments for both generations 1 and 3 in relevant production traits. Furthermore, microbial analysis is currently ongoing to investigate if the difference in performance is due to differences in microbiomes. The presentation will showcase results for measured traits alongside microbiome composition of BSF eggs, juveniles, and harvest larvae from both parent and interchanged populations. This study has implications for understanding adaptation responses of BSF to low-quality diets and for optimizing the production of insects.

Session 53

Poster 20

Effect of broodstock breeding density on mortality and reproductive performances of *Tenebrio molitor* (L.)*B. F. Palumbo¹, M. Cullere¹, Y. Singh¹, E. Pontalti¹, A. Dalle Zotte¹**¹ University of Padova, MAPS, Viale dell'Università 17, 35020 Legnaro, Italy*

The study investigated the impact of 4 different broodstock breeding densities (0.8, 1.1, 1.3 and 1.6 breeding animals/cm², D1, D2, D3 and D4, respectively) on mortality and reproductive performances of yellow mealworm (*Tenebrio molitor*). Broodstock were randomly assigned to the 4 groups and housed in 48 breeding crates (60 × 40 × 14.5 cm; 12 crates per group). The trial consisted in a total of 4 weeks of broodstock maintenance (4 oviposition batches). Body weight (BW) and mortality of broodstock, as well as the hatching rate of eggs were assessed once a week; ovaries were collected, and their proportion on the total body weight of the broodstock assessed at 4 weeks of age. The number of larvae per crate was estimated at 5 weeks of age; larvae BW was assessed at 5 and 8 weeks of age. Data were analysed by two-way ANOVA with breeding density and week of breeding/batch of larvae as fixed effects by using the GLM procedures of SAS. Mortality of broodstock tended to increase from 20.5% (D1) to 22.4% (D4; $P=0.054$); however, no significant differences were observed among the breeding densities in terms of hatching rate, which averaged 85%. The ovary proportion was higher in D2 compared to D1 ($P<0.05$), whereas the highest BW was observed in D4 and the lowest in D2 ($P<0.05$). The number of larvae/crate significantly increased ($P<0.001$) whereas the number of larvae/adult linearly decreased with increasing adult breeding density ($R^2=0.85$). The present study demonstrated that a high broodstock breeding density in yellow mealworms can increase the mortality rate while decreasing the reproductive performances in terms of produced larvae.

Effects of light regime and starvation on immune parameters in house crickets (*Acheta domesticus*)

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Crickets are a protein source with a lower imprint on the environment than traditional livestock. However, food safety is a main issue and a common way to reduce the microbial load is fasting the insects for 24–48 hours. This may pose a physiological stress for many insects and could indicate a welfare problem. Another parameter that can be altered in cricket rearing is the light regime, which affects maturation but may also affect immunity. In this study, the aim was to investigate the combination of these two parameters on cricket immune response parameters. Male and female house crickets (>4 weeks post-molt) were reared at 30 °C (±1 °C), 50% relative humidity (±10%) with access to shelter and ad libitum water, using a 16L:8D or 12L:12D light regime. Before euthanasia they were starved for 48 hours or fed ad libitum. Crickets were euthanized by freezing, hemolymph was extracted, diluted and used for measuring phenoloxidase (PO), protein content and hemocyte count (n=4–9 for all groups). Data were analyzed using a 3-way ANOVA with light regime, starvation or ad libitum feeding and sex as categorical factors. P<0.05 was considered significant. Main effects were seen on PO-activity (p=0.002) and hemocyte count (p=0.005), both lowered by starvation, suggesting a possible effect on immunocompetence. A 16L:8D light regime lowered the protein content in hemolymph compared to 12L:12D (p=0.045), which by itself is not an immune parameter but have been correlated to disease resistance in previous studies. The results point towards starvation as a non-sustainable method from a welfare perspective but need to be replicated and investigated further to make recommendations for future cricket rearing.

Session 54

Theatre 1

Exploring Antibiotic Alternatives for Improved Health in Goat Kids

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The emergence of and potential for antibiotic resistance poses a significant threat to global public and animal health, making the exploration and development of alternative strategies to combat bacterial infections imperative. The overuse and misuse of antibiotics in livestock production have accelerated the development of resistant bacterial strains in both animals and humans. Commercial goats are raised globally for meat, milk and fibre purposes in a variety of production systems including extensive grazing and intensive housed conditions. The health of goat kids, particularly in intensive production systems, can be challenging and potentially compromised by infectious diseases, leading to substantial economic losses and welfare concerns. Prewaning mortality in goat kids is estimated to range from 6% to 38%, commonly caused by similar issues found in dairy calves including pathogens (e.g. *Escherichia coli* and *Staphylococcus aureus*), gastrointestinal, and respiratory diseases. We recently showed that although metaphylactic antibiotics can halve preweaning mortality, similar improvements are likely to be achieved via increased successful transfer of passive immunity (STPI) rates. Our findings show that STPI can triple survival rates, improve growth performance and enhance average daily gain in dairy goat kids. This review will summarise the current knowledge regarding antibiotic alternatives for goat kids including colostrum feeding duration and methods, probiotics and prebiotics, plant-based products, yeast and fermentation products and essential oils.

Sustainable Sheep Milk: Exploring Udder Health Traits and Antioxidant Activity

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Limited understanding exists on exploiting antioxidant activity in milk from dairy ewes due to challenges in gathering data from numerous samples. Obtaining individual data over time is essential to characterize local populations and understand milk quality variability. This study aims to investigate the effect of breed, parity, lactation, milk gross composition, udder health traits, and minerals on the total antioxidant activity in sheep milk, using a General Additive Mixed Model. A total of 720 individual ewe milk samples were collected from Comisana and Massese breeds at the nucleus breeding center of the Italian Sheep and Goat Breeders Association. The total antioxidant activity was assessed via an electron-transfer technique (DPPH; % inhibition) and a ferric reducing antioxidant power (FRAP; μM) assay. Massese and Comisana breeds showed average values of 63.0 ± 2.2 and 63.6 ± 2.3 and 2.24 ± 0.51 and 2.23 ± 0.55 for DPPH and FRAP, respectively. Milk composition highly affected FRAP while DPPH was marginally influenced. These findings can improve knowledge on the nutraceutical and technological properties of sheep milk. They also serve as a tool to ensure breed conservation, maintain traditional farming systems, and increase farmers' profits. The results are embedded within the research contract of Giorgia Stocco, co-financed by the European Union – NOP Research and Innovation 2014-2020 art. 24, par. 3, A) Law 30/12/2010, n. 240 and of the D.M. 10/08/2021 n. 1062.

Session 54

Theatre 3

Changes in lactating dairy goat body composition

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The ability of an animal to retain and mobilise body tissues is important for determining lifetime productivity. This study investigated changes in body composition in high (HP) and low (LP) producing commercial dairy goats utilizing non-destructive assessment techniques including Dual Energy X-Ray Absorptiometry (DEXA), body weight (BW), body condition score (BCS), and sternum girth. Saanen dairy goats (20 HP and 20 LP) were used. On four occasions, early (EL, 23 ± 6 DIM), mid (ML, 107 ± 6 DIM), late (LL, 206 ± 6 DIM) and 2 weeks post drying off, goat body composition was measured (BCS, BW and sternum girth by caliper). During DEXA (Horizon W) scans, goats were placed in a prone position with back legs extended and front legs back beside the trunk. DEXA scans were analysed by regions (whole body (excluding head); WB and sternum region (from 1st-6th sternal rib). Statistical analysis performed using GenStat (22nd Edition) using linear regression and REML mixed models. By design, HP had higher milk yield than LP overall (2.6 vs. 2.2 L/d; $P=0.004$). BW, BCS, and WB lean muscle mass increased, while WB fat decreased from EL to ML and ML to LL ($P<0.001$). Sternum lean muscle and girth increased from EL to ML and ML to LL, whilst sternum fat % decreased ($P<0.001$). There were no differences between HP and LP goats. Sternum girth was not correlated with sternum fat or WB fat, indicating that sternum girth is not a reliable predictor of body fat, but is moderately correlated with BCS ($R^2=0.59$; $P<0.001$). This suggests BW, BCS and sternum girth are not reliable indicators of fat distribution or change in dairy goats.

Effect of the replacement of wheat straw by spent mushroom substrate on milk yield, composition, fatty acid profile, oxidation stability and udder health in dairy ewes during the final stage of lactation
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The aim of the present study was to examine the effects of different levels of spent mushroom substrate (SMS) at the expense of wheat straw (WS) on milk yield, composition, fatty acid profile, oxidative stability and udder health in sheep. Thirty ewes at their final stage of lactation were randomly allocated into 3 groups; the first one served as control (C) and fed with a diet consisting of concentrates, alfalfa hay and WS, whereas in the other two groups WS was replaced by SMS at 50 or 100% (SMS1 and SMS2, respectively). The experiment lasted for 28 days, and milk yield, composition, oxidative stability and somatic cell count (SCC) were weekly monitored, while milk fatty acid and immune cell profile were also determined on day 28. Data were subjected to ANOVA with SMS treatment as fixed effect. The linear or quadratic dose response to SMS were tested with polynomial contrasts. The significance level was set at $P=0.05$. Statistical analysis was applied with the SAS software. No significant differences were observed in milk yield, composition, SCC and fatty acid profile among the experimental groups. However, milk oxidative stability was significantly improved as an effect of SMS ($P<0.001$). At the same time, polymorphonuclear leukocyte percentage was decreased in SMS2 group ($P<0.05$). SMS seems to be a promising agro-industrial by-product for ewes' diet that could improve milk oxidative stability, without negatively affecting milk yield, composition and ewe health status. * This research project was implemented within the framework of the Project "Residues2value", code: ATTP4-0339570, MIS 5185063, Funding Body: Hellenic State & European Union

Camelina and cardoon cakes supplementation on blood metabolic profile and colostrum quality in dairy goats during the transition period

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The study aimed to evaluate the impact of camelina (*Camelina Sativa*) and cardoon (*Cynara Cardunculus*) cakes on metabolic blood profile and colostrum quality of transitioning dairy goats. 36 Alpine multiparous dairy goats (age: 3.04 ± 0.85 years; milk yield: 3.93 ± 1.23 L/day) were individually housed and randomly assigned into four isonitrogenous and isoenergetic diets ($n=9$): a control diet (C), without experimental supplementation; a diet supplemented with 4.98% of cardoon cake (CD); a diet supplemented with 5.92% of camelina cake (CAME); a diet supplemented with 5.23% of a mixture of cardoon and camelina cakes (CACD). The trial lasted from 21 days prepartum to 21 days post-partum. Individual feed intake and milk yield were recorded daily. Colostrum samples were collected immediately after kidding while blood samples were collected at day -21, 2 and 21. Colostrum functional profile was assayed using the Folin-Ciocalteu method for quantification of total phenolic content and the FRAP (Ferric Reducing antioxidant Power) assay for determination of antioxidant activity. Performance and blood data were analyzed by mixed procedure of SAS for repeated measurements, colostrum data were analyzed by one-way Anova. Significance was set up for $p<0.05$. No treatment effects were observed on feed intake, milk production and main blood metabolites. At 21 days of trial blood urea tended to be higher in CACD group ($0.10<p<0.05$). CAME, CD and CACD colostrum showed higher total polyphenols compared to C (388.3 ± 17.6 ; 395.42 ± 12.99 ; 396.88 ± 20.10 vs 316.20 ± 14.58 \square g TAE/mL; $p<0.05$). CACD colostrum had higher antioxidant capacity compared to other groups (80.51 ± 3.98 vs 40.77 ± 2.97 ; 59.68 ± 6.58 ; 58.35 ± 6.31 \square g AAE/mL; $p<0.05$). Overall, the supplementation with CAME and CD alone and in mixture seems to improve the quality of colostrum, without negatively impact performance and health status of dairy goats.

Impact of somatic cell score on milk composition, curd firming, and cheese-making traits of Manchega sheep

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The aim of this research was to assess the impact of somatic cell score (SCS) on different phenotypes related to production, composition, milk colour, traditional milk coagulation properties (rennet coagulation time – RCT –, curd-firming rate as the time to a curd firmness – CF – of 20 mm, and CF at 30 and 60 min of analysis), and new curd firming and syneresis traits (modeled RCT, potential asymptotical CF at an infinite time, curd firming and syneresis instant rate constants, maximum CF value and achievement time); cheese yield traits, as the weights of total fresh curd, dry matter, water in the curd and nutrient recovery traits. The study included individual milk samples from 791 Manchega ewes reared in 5 flocks. All these phenotypes were analysed using a mixed model that considered, in addition to the standard nuisances, the effect of SCS discretized into 7 classes of half standard deviation. A significant effect of SCS was observed on almost all traits. In addition to the expected results on standard traits, findings related to milk colour, curd firmness, and cheese characteristics were particularly interesting. Specifically, as SCS increased, milk lightness decreased and a^* values approached red. SCS also had a negative effect on coagulation, delaying milk gelation and reducing CF. Furthermore, curd moisture increased with SCS and there was lower recovery of protein in the curd. This research is part of Project PID2020-118031RR-C21/AEI/10.13039/501100011033.

Session 54

Theatre 7

Mammary gland modulation on metabolic and immune status of dairy goats and their offspring

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Neonatal mortality has become an important challenge for the dairy goat industry. Newborn ruminants are highly dependent on good quality colostrum intake right after birth to obtain energy and an appropriate immunization. Colostrum quality is mainly determined by IgG concentration, however other less abundant molecules such as lactoferrin or oligosaccharides are crucial for the immunization of the newborn goat kid. Several factors, such as dam nutrition or udder health during late gestation play a relevant role on colostrum quality. Dam metabolism can be affected by high starch diets during parturition which induce changes in blood metabolites (i.e., glucose, BHB and FFA) and colostrum bioactive compounds (i.e., insulin). In addition, several studies have assessed the effects of mastitis on physiological responses within the mammary gland. For instance, the intramammary administration (IA) of immunomodulatory molecules such as lipopolysaccharides (LPS) from *Escherichia coli* (O55:B5) at parturition has shown not only to increase colostrum quality (i.e., increased IgG and IgM concentrations) without having negative effects on dairy goats but has also enhanced the immunization of those newborns receiving colostrum from LPS challenged goats. These results suggest that dam metabolism and mammary gland physiology can be modulated by parturition diets and the application of molecules such as LPS. However, further studies are necessary to determine the suitability of these strategies to improve colostrum quality in practical conditions.

The role of colour indexes for the assessment of species-specific and breed-specific differences in quality traits of milk from domestic ruminants

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This work presents results from a line of research based on the study of colour of milk from domestic ruminants, aiming to assess if colour can be used as a discriminator, not only at species level, but also at breed level. Additionally, we seek to explore if colour can be used to predict technological quality of milk intended for cheesemaking. For this purpose, over 3200 milk samples from sheep, goats and cows were analysed. Laboratory analysis included milk composition (obtaining values for fat, crude protein, lactose and total solids) and milk coagulation properties (including traits such as rennet clotting time – RCT, curd firming time – k20, curd firmness at 60 min. –A60, curd yield – CY, and curd yield solids – DCY). Colour indexes of milk were expressed using the CIELAB colour space as three variables: Lightness (L*), green-red balance (a*) and blue-yellow balance (b*). By combining discriminant analysis, cluster analysis and canonical correlation analysis, differences and similarities at a species and breed level were identified, and covariation patterns between the variables described above were evaluated. All statistical analyses were performed with XLSTAT v.19.4 (Addinsoft, New York, NY, USA). Findings revealed that colour can be used to effectively discriminate milk at a species level and, to some extent, at a breed level. While milk quality traits show common patterns of variation, strong species-specific and breed-specific relationships emerge, highlighting the connection between milk colour and curd yield. This could allow to model these relationships in future studies to estimate milk technological performance, potentially leading to an optimization of the coagulation process. In addition, measurement of colour in milk acquires great interest as a rapid and inexpensive tool to somehow predict milk quality.

Session 54

Theatre 9

Impact of reproduction on longevity of ewes

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Modern selection on ewes has focused mainly on production output, increasing litter size and autumn weight. In contrast to other countries, it has been common to mate ewe lambs in Norway since the 1970's. This is to increase the lifetime production of ewes. However, no one has investigated how the practice affects the lifespan of the ewes. We used data on Norwegian White Sheep spread across Norway to investigate the effect of age at first lambing on the lifetime reproduction and lifespan of the ewe. We used linear mixed models and the lme4-package in R with herd x year as a random effect. Non-significant effects were excluded. We included culled ewes that stayed in the same herd their entire lifetime. Ewes were excluded if they did not rear the same number of lambs with the same gender ratio as they lambed in all parities, and if a parity was longer than 3.5 years. A total of 548947 ewes born between 2000 and 2020 were included in the final analysis. They were split into three groups according to age at first lambing (1, 2 or 3 years old). The groups included respectively 77% (1), 21% (2), and 2% (3) of the ewes. The average lifespan was 3.27 years (1) and increased by respectively 0.84 years (2) and 1.77 years (3). The average number of weaned lambs at 90 days, summed over the lifespan was 4.31 (1 and 2) while there was a drop of 0.27 lambs for ewes in group 3. Ewes wean the most lambs when they are 4-5 years old. This shows that ewes that have their first lambing delayed live longer, but not long enough to compensate for the non-reproductive period, and that ewes that lamb when they are 1 year old often are culled before they reach their maximum reproductive potential.

Assessment of mineral composition in sheep milk: which effects should we include?

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Among the various components that contribute to sheep milk's appeal and value are minerals, which maintain both the milk's structural integrity and nutraceutical qualities. It has been known that milk composition varies due to factors such as breed, genetic variation within breed, health, environment, and farming practices. However, limited research has been conducted on exploring additional factors that can affect mineral composition in sheep milk. Individual milk samples from 429 Comisana (CO) and 311 Massese (MA) reared at Asso.Na.Pa nucleus center were collected once during morning milking and analyzed for milk composition and mineral profile. A linear mixed model, including days in milk (DIM), parity order (PO), sampling date, and milk characteristics as fixed effects, and animal as random effect, was used to study minerals variability. The analysis revealed a significant effect ($P > 0.05$) of casein, lactose, and SCS on K, Cl, Na, Mg, and S. Additionally milk pH and fat significantly affected Ca, K, and Cl. Milk pH and SCS were also important on S. The results of this investigation imply that variables including pH, lactose, and casein might also have an impact on the mineral composition of sheep milk. When evaluating the total mineral content, these elements need to be considered.

Could morphological traits assist towards increasing goat milk productivity in Cyprus?

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Damascus goats and their milking capacity in particular, are of pivotal significance for the Cyprus milk-related industry as well as for achieving the Halloumi milk-origin requirements as a Cyprus protected destination of origin (PDO) product. In order to contribute to the high demand for goat and sheep milk, through this study, we focused on the development of a novel morphological database for Damascus goats in Cyprus, covering quantitative and qualitative traits with emphasis in udder traits. Udder morphological traits were recorded in 2024, during their non-lactation and lactation period (2 weeks post-partum and 2 weeks post-weaning) thus allowing the evaluation of the morphological effects at three different udder stages (relaxed, post-partum, and post-weaning) on milk production. Individual goat milk production was documented daily by an automated milking device. By using a multi-model inference approach, we evaluated the effects of each trait and stage, towards the milk production capacity for each individual. Based on the weighted significance of each morphological character, we proceeded in the development of the best possible (stage-wise) morphological index associated with the milk production. Subsequently we aim, through long-term trait documentation to search for age correlations regarding the significant traits, thus allowing the use of the morphological index as an early age predictor of goat milking potential.

Effects of age at first joining and genotype on ewe performance from 2 to 8 years of age

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The objective was to evaluate the effect of first-joining age (~7 or 19 months) and genotype on ewe performance from 19 months until aged 8.5 years. A total of 424 ewes [Belclare (B), Belclare×Suffolk (BS), Suffolk type (>75%S); 2 cohorts] were joined with Charollais rams each year. A random 50% of each genotype had been joined at 7 months, of which 86% lambed and 1 lamb was reared per ewe joined. Ewes exited the flock when culled for health/welfare reasons (e.g., udder issues) or they died. Ewes lambed indoors (March) and were put to pasture within 3 days. Ewes rearing triplets received concentrate (0.5 kg/d) for 5 weeks post lambing, and their lambs had access to concentrate (max 300 g/d each) until weaning (14 weeks). Between weaning and slaughter all lambs co-grazed on pasture (sole diet). The data were analysed using Proc MIXED of SAS. There were no interactions between first-joining age and genotype ($P > 0.05$). Litter size, lambs reared/ewe joined, lamb mortality, number of joinings and lamb weaning weight (kg) for the first-joining treatments (7 and 19 months) were: 1.96 and 1.95 (NS); 1.58 and 1.53 (NS); 6.7 and 9.2% ($P < 0.05$); 3.9 and 3.7 (NS); 33.4 and 33.2 kg (NS), respectively. The corresponding variables for B, BS and >75%S ewes averaged: 2.02, 2.09 and 1.76 ($P < 0.001$); 1.62, 1.70 and 1.34 ($P < 0.001$); 6.1, 6.8 and 11.3% ($P < 0.05$); 3.6, 4.0 and 3.8 (NS); 33.0, 34.1 and 32.8 kg ($P < 0.01$), respectively. Total lambs reared/ewe over the duration of the study was 6.2 and 5.7 (NS) for the first-joining at 7 and 19 months groups (NS). Corresponding means for B, BS and >75%S were: 5.9, 6.8 and 5.2 ($P < 0.01$). When the lamb output from joining at 7 months is accounted for, this treatment yielded an additional 1.5 lambs reared per ewe lifetime: similar to the response from changing ewe genotype.

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Poster 13

Doses of chromium propionate in lamb diets and their effects on metabolic parameters

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Chromium is an essential micronutrient in lipid and glucose metabolism, and its trivalent form, CrIII, enhances insulin signaling. Organic sources of chromium, such as chromium propionate, can be up to ten times more bioavailable than inorganic sources and have an effect on metabolism. This study was designed to evaluate the metabolic effects of two additional doses of chromium propionate. 29 uncastrated male lambs, 90 days old and weighing an average of 21.16 kg, were used and divided into 3 treatments: CTL, no added chromium; Cr0.5, 0.5 mg/kg DM; and Cr1.5, 1.5 mg/kg DM. The experimental period lasted 64 days, during which time blood samples were taken for biochemical analysis. The experimental design was completely randomized, and means were compared using the Tukey test at 5% probability. Cholesterol ($P=0.0023$) and phosphorus ($P=0.0213$) were lower in Cr0.5, and alkaline phosphatase decreased linearly ($P=0.0274$). Cr improved the parameters of total leukocytes ($P=0.0126$), lymphocytes ($P=0.0476$) and monocytes ($P=0.0058$). Chromium acted on lipid metabolism by reducing cholesterol and with indications from the lymphatic system of less inflammation. The chromium diet interacts with phosphorus, which consequently alters the value of alkaline phosphatase, but all the values were within the reference values for sheep. Chromium propionate supplementation at 0.5 mg/kg DM for lambs was beneficial for metabolism and the immune system.

Does dairy sheep breed influence the environmental impact of farm?

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The global livestock sector contributes a 14.5% to anthropogenic greenhouse gas emissions with small ruminants accounting for 596 million tons of CO₂ eq. In Tuscany (central Italy) dairy sheep breeding enterprise is an important sector and the breed mainly reared are the Lacaune (L), the native Sarda (S) and Massese (M). The aim of this study was to apply the Life Cycle Assessment (LCA) methodology to carry out a descriptive analysis of the environmental impacts of sheep milk production in three farms in Tuscany, similar in terms of rearing system and flock size, but differing by the breed reared (L, S and M). LCA method was applied according to the ISO 14040-44. The selected system boundary was "from cradle to farm gate" and the functional unit 1 kg of fat and protein corrected milk calculated with the formula of the International Dairy Federation (2015). A biophysical allocation based on the energy requirement for milk and meat production was applied following the FAO's LEAP guidelines (2016). Primary data were collected on site, while secondary data were taken from Ecoinvent 3.9.1 database. Environmental impacts were calculated with the EF 3.1 method through the OpenLCA software (1.11 version) and the impact categories studied were: climate change, freshwater, marine and terrestrial eutrophication, acidification, water and land use. The L farm showed the lowest values for most of the impact categories, and the M the highest. These results were due to the higher productivity of the L breed compared to the other two breeds. The M farm showed the lowest impact only for the water use. These results suggest that further studies should evaluate the impact taking into account the breeds and integrating, at the one time, other aspects such as the protection of biodiversity or the role of ecosystem services provided by the livestock.

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Poster 15

Analysis of fattening traits of black-headed lambs in Poland in 2015-2020 assessed by the BLUP method.

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The black-headed breed is one of the most important paternal breeds of sheep in Poland, used for commercial crossbreeding to improve fattening traits of lambs. Genetic evaluation using the BLUP method is an effective selection tool for estimating the breeding value of animals and improving the population. The aim of the study was to analyze the genetic progress of daily gains and body weight of black-headed lambs in Poland in 2015-2020. The study was conducted on the basis of data on the breeding value of 9 715 lambs (4698 rams and 5017 ewes) of the black-headed breed, collected as part of the genetic evaluation conducted by the NRIAP in Krakow in the years 2015-2020. The analysis included daily gains from 10-30 and 30-56 days of age and body weight at day 56. The results were statistically processed with the STATISICA ver. 10 package [13] using one-way analysis of variance. Testing was carried out at the significance level of $p \leq 0.05$ and $p \leq 0.01$. When a significant effect of a factor on the studied trait was found, Duncan's chisquare test was used. A gradual improvement in the breeding value of black-headed lambs for fattening traits was observed between 2015 and 2020. The average breeding progress for daily gains was 2.5% per year and for body weight at day 56 – 1.8%. During the analyzed period, average daily gains between 10-30 days varied: for rams from 241 g to 280 g, for ewes from 233 g to 267 g. On the other hand, average daily gains between 30-56 days: for rams from 242 g to 291 g, and for ewes from 241 g to 281 g. Body weight for rams at 56 days of age, during the period analyzed, ranged from 18.2 kg in 2015 to 19.5 kg in 2020, and for ewes 17.2 kg in 2015 and 18.8 kg in 2020, respectively. Genetic evaluation using the BLUP method enables improvement of daily gains in rams and ewes of the black-headed breed. The rams show higher gains and faster breeding progress than the ewes, especially in the later period of rearing.

Effect of rearing sucking lambs in mountaing areas on carcass quality and meat fatty acid profile

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Extensive livestock systems are of great importance for the conservation of biodiversity. Sheep grazing on natural mountain pastures and rearing lambs that are marketed under the brand "Ganadería Probiodiversidad" contribute decisively to the conservation of the habitat of the bearded vulture, an endangered scavenger bird. Meat from these lambs could be differentiated in the market by unique characteristics. The aim was to evaluate the effect of the rearing system (naturally suckling lambs reared on mountain pastures, BIO, versus suckling lambs reared on milk replacer, MR) on carcass and meat quality. Ten carcasses from each type of production were measured and assessed for fatness and colour. Afterwards, longissimus muscle was removed for chemical composition and fatty acid profile analysis. Data were subjected to one-way anova. No differences were observed in carcass weight (7.5 kg) and meat proximate composition (average 72.0% moisture, 19.4% protein and 4.3% fat content). However, BIO carcasses were wider and had higher muscle colour intensity, whereas BIO meat showed high percentages of PUFA, particularly linoleic (1.48 vs. 0.43) and conjugated linoleic acid (1.65 vs. 0.44) and a more favourable n-6/n-3 ratio (2.2 vs. 10.0, for BIO and MR, respectively). Therefore, in addition to benefits of production system on the environment, natural suckling lamb reared with mothers in mountain areas, showed high intense colour, and more favourable fatty acid composition, in accordance with health recommendations on meat consumption. Work funded by the Biodiversity Foundation (MITECO), Recovery, Transformation and Resilience Plan framework, financed by the European Union – NextGenerationEU.

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Poster 17

Effects of Cysteine on Secondary Hair Follicle Growth and Hair Dermal Papilla Cell Proliferation in vitro in Yanshan Cashmere Goats

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The aim of this study was to investigate the effects of cysteine (Cys) on the growth of secondary hair follicles (SF), the proliferation and apoptosis of hair dermal papilla cells (DPCs) cultured in vitro of Yanshan cashmere goats. The skins of one healthy male Yanshan cashmere goat at one year old was selected, and the completely isolated secondary hair follicles were cultured for 7 days with the concentrations of the Cys at 0, 40, 80, 120, 160, and 200 $\mu\text{g}\cdot\text{mL}^{-1}$, respectively. The hair follicle growth pattern was observed and hair follicle length was measured every 24 h. DPCs were extracted and purified from the secondary hair follicles, and cultured with the concentrations of Cys at 0, 20, 40, 60, 80, and 100 $\mu\text{g}\cdot\text{mL}^{-1}$, respectively. Cell proliferation viability was detected using the CCK-8 kit, and the qRT-PCR was used to detect the mRNA expression levels of proliferation-related genes (PCNA, CCND1, CDC42, CDK4), apoptosis-related genes (P21, P53, Bax, Caspase-3, Bcl-2), skin cell differentiation-related gene (IVL) and keratin-related genes (K10, K14). All statistical analyses were carried out using SPSS version 22.0. Data were analyzed by one-way analysis of variance (ANOVA) and multiple comparisons among groups were performed using Fisher's protected least significant difference (LSD) test. All data are presented as mean \pm standard error of the mean (SEM). P-value < 0.05 was considered to indicate statistical significance. The results indicated that the addition of 80 and 120 $\mu\text{g}\cdot\text{mL}^{-1}$ Cys significantly affected the growth rate and cumulative growth length of secondary hair follicles ($P < 0.05$). The addition of 40 $\mu\text{g}\cdot\text{mL}^{-1}$ Cys significantly affected the proliferation of DPCs ($P < 0.05$), and significantly up-regulated the mRNA expressions of PCNA, CCND1 and CDK4, Bcl-2, K10, K14, and IVL gene. In conclusion, the appropriate addition of Cys could promote the growth of secondary hair follicles and the proliferation of hair dermal papilla cells in vitro through the regulation of genes for cell proliferation, differentiation and keratin in Yanshan cashmere goats.

Diurnal variation in circulating blood metabolites in lactating dairy goats

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Understanding diurnal variation in key metabolites such as non-esterified fatty acids (NEFA), glucose and blood urea nitrogen (BUN), is important for monitoring health status and determining a reference range in dairy goats. This research aimed to investigate differences in the diurnal variation in circulating blood metabolites over a 48-hr period in high (HP) and low (LP) producing dairy goats. 29 Saanen early lactation dairy goats (15 HP and 14 LP) were housed individually for 5 days and fed TMR (1.8x maintenance) twice daily. Blood samples (every 2hrs over 48hrs) were collected via a jugular catheter. Plasma was harvested and stored at -20°C prior to metabolite analysis using validated commercial kits. Statistical analysis was performed using GenStat (22nd Edition) using REML mixed models. HY goats had greater milk yield than LY goats (3.9 vs. 2.2 L/d; $P < 0.001$). Average (over 48h) concentrations of blood glucose and NEFA were lower for HY compared to LY (2.7 vs 3.1mM; $P = 0.04$; 658 vs 323µM; $P < 0.001$, respectively) whilst BUN did not differ between production groups. Glucose, NEFA and BUN differed across all time points ($P < 0.001$). Glucose was lower in AM, whilst NEFA and BUN were highest in AM. These findings indicate that early lactation dairy goats display diurnal variations in circulating blood glucose and NEFA. Moreover, HY goats have lower blood glucose and higher NEFA compared with LY goats, but no differences in BUN. Identification of the natural patterns of key metabolites and differences between production levels will assist future research in establishing appropriate blood sampling time points and reference ranges for key metabolites.

Session 54

Poster 19

Lambs fed Kraft lignin, ionophore and vitamin e with selenium on performance, meat quality and rumen health

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Kraft lignin is a byproduct of the alkaline hydrolysis of wood that contains low molecular weight phenolics that have antioxidant activity. Thirty-two lambs of the Dorper and Santa Inês breeds, aged 90 days and 20 kg BW, were confined in individual stalls and equally distributed in four treatments: diet without additives (CTL), diet with lignin (Lig), diet with monensin, selenium and vitamin E (Mon +SeE), and diet with lignin, monensin, selenium and vitamin E (LigMonSeE). The concentration of lignin was 18 g/kg DM, monensin sodium (Rumensin®Elanco) was 16 mg/kg DM, selenium was 0.33 mg/kg and E vitamin 100 IU/kg DM. The animals were fed for 60 days and weighed on the first day and then at 15-day intervals. The data obtained was analyzed using the SAS with the MIXED procedure and the Tukey test at 5% statistical probability. The treatments had no effects on animal performance and carcass characteristics ($P > 0.05$) with an average final weight of 38.87 kg. Daily weight gain, feed conversion and dry matter consumption did not differ between treatments, ranging from 0.25 to 0.31 kg, from 4.36 to 5.09 kg/kg, and from 3.19 to 3.45%, respectively. Cold carcass yield, loin eye area and subcutaneous fat thickness showed values ranging from 44.12 to 45.13%, 14.78 to 16.52cm² and 2.03 to 2.49mm, respectively. However, the animals fed with MonSeE had more visceral fat than the other treatments ($P < 0.05$). There was no significant rumen inflammation. It can be concluded that the purified Kraft lignin did not adversely affect performance, carcass quality and rumen health.

Particle size distribution of total mixed rations in Spanish Assaff dairy sheep farms

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In order to characterize the particle size distribution (PSD) of total mixed rations (TMRs) and to evaluate the relationship of the physically effective fiber (peNDF) with milk production and composition, 14 commercial Spanish Assaff dairy sheep farms were selected. On each farm, three high-producing TMR samples were taken along the feed bunk and PSD was determined using a Penn State Particle Separator (PSPS). Chemical composition of the TMRs was analyzed and milk production and composition was recorded. Of the 14 farms, 9 administered silages (TMR-S) and 5 administered hay (TMR-H) as the predominant forage base. Statistical analyses were performed using SAS package. Results were analysed by GLM procedure including TMR type as a fixed effect. Simple correlations were determined for NDF, peNDF, PSD and milk production and composition using CORR procedure. The statistical significance of differences was defined as $P < 0.05$ and trends as $P < 0.10$. As expected, DM content was lower in TMR-S than TMR-H ($P < 0.001$, 62.0% vs. 84.7%). No statistical differences ($P > 0.05$) were found in chemical composition and PSD between TMR-S and TMR-H. The mean particle retention above the 19 mm, from 8-19 mm, from 4 to 8 mm and less than 4 mm were 15%, 27%, 25% and 34%, respectively. Within each farm, the highest mean value of the coefficients of variation (34%) was for > 19 mm fractions, while the minimum (12%) was for the 8-19 mm fraction. A negative correlation between peNDF > 8 mm ($P < 0.05$, $r = -0.57$) and peNDF > 4 mm ($P < 0.05$, $r = -0.61$) was found with milk production. Milk fat content tended to be positively correlated ($P = 0.10$, $r = 0.47$) with particles between 8-19 mm. The TMRs studied were rich in long particles and most of them poorly mixed. Further controlled studies are required to establish specific PSD recommendations of TMRs for dairy sheep and to ensure a well-mixed rations that prevents diet selection and supports rumen function, animal welfare and optimal performance. Grant PID2020-113395RB C22 funded by MCIN/AEI/10.13039/501100011033.

Session 54

Poster 21

Doses of chromium propionate on the performance of finishing lambs

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Chromium is an enhancer in energy diets to improve animal performance, but the recommendation for chromium in ruminants has not been established. This study aimed to evaluate the effects of two supplemental chromium propionate doses on the performance of finishing lambs. A total of 29 uncastrated male lambs, age of 90 days, with an average weight of 21.16 kg, were divided into 3 treatments: CTL, no added chromium; Cr0.5, 0.5 mg/kg DM; and Cr1.5, 1.5 mg/kg DM. The experimental period lasted 64 days, the animals were weighed to assess performance, and individual and daily feed consumption was monitored. The experimental design was completely randomized and means were compared using the Tukey test at 5% probability. The inclusion of chromium propionate increased dry matter intake ($P < 0.0001$), which was 1.27, 1.29, and 1.48 kg per day for the CTL, CR0.5, and CR1.5 treatments, respectively. However, there was no difference in average daily weight gain, which averaged 0.384 kg/d. The lambs were slaughtered at an average weight of 44.25 kg and there was no difference between treatments. As a result, there was a lower feed efficiency for the CR1.5 treatment with a value of 0.26 kg/kg, and no difference between the other treatments with a value of 0.31 kg/kg. It can be concluded that the concentration of 0.5 mg/kg DM was similar to CTL for the parameters studied, but the supplementation of 1.5 mg/kg DM worsened the performance of the animals.

Manure management in dairy sheep farms

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The aim of this study was to explore the practices of dairy sheep farmers in Castilla y León (Spain) regarding management of the manure. To do this, a questionnaire about the on-farm manure management practices was filled by the researchers and in the same visit information about bedding state, ewe cleanliness and ammonia level was collected in 20 dairy sheep farms. Data set were analysed using the non-parametric Kruskal-Wallis test and CORR procedure of SAS 9.4. software. Of the farms surveyed, 40%, 30% and 30% had flock sizes of < 700 ewes (1), 700-1400 ewes (2) and > 1400 ewes (3) respectively. All farmers use straw as bedding materials, and straw was added periodically as required to maintain adequate bedding conditions. 50% of farms added straw daily, 40% every two days, while 10% every three days. In all farms that added straw daily the bedding state was considered appropriate. However, when straw is added every two or three days, 25% and 50% of the farms respectively had an inadequate bedding state. A positive correlation ($P < 0.001$, $r = 0.80$) was observed between the bedding state and the cleanliness of the ewes. None of the farms had obvious ammonia odors or eye irritation and 20% of the farms did not have any ammonia odors. Eighty per cent of the livestock farms removed manure at least every two months. More than half of the farms stacked manure into piles above-ground, of which 45% are type 1 farms. On the other hand, 25% do not store manure, of which 60% are type 3 farms. The size of the farm only tended to affect the cleanliness of the ewes ($P < 0.10$), with larger farms showing a better assessment of ewe cleanliness. These results could help to establish the optimum manure management in Spanish dairy sheep farm. Grants PID2020-113395RB C22 funded by MCIN/AEI/10.13039/501100011033.

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HereForEwe: Implementation of sheep production system of minor dairy breeds through community-based breeding, genomics and phenomics: the example of Delle Langhe breed

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Small ruminant breeding is vital for rural development. However, European ovine milk production declined due to competition from dairy cattle. In Italy, 75% of sheep breeding focuses on dairy products, thus facing profitability challenges. Delle Langhe breed, once economically important, is now facing challenges to be sustainable due to the loss of market relevance and genetic diversity. Research projects of national interest funded by the European Union (EU) such “HereForEwe” (NextGenerationEU, code 2022HJJNB5), aims to highlight the peculiarities of the breed, considering milk quality, disease resistance and genetics. Thorough milk composition, coagulation traits and infrared spectra will be collected on over 1,200 ewes. These will undergo SNP chip genotyping to confirm pedigree, estimate inbreeding, identify favorable alleles for milk traits and disease resistance and to reveal lethal or unfavorable haplotypes. Identification and validation of miRNAs as disease biomarkers for stress resilience pathway exploration will also be performed. The “HereForEwe” project aims to engage breeders in community-based breeding programs for effective genetic improvement aligned with owner preferences, by addressing challenges faced by local breeds. Part of the results are embedded within the research contract of Giorgia Stocco, co-financed by the EU – NOP Research and Innovation 2014-2020, art. 24, par. 3 a), of Law 30/12/ 2010, n.240 and subsequent amendments and of the D.M. 10/08/2021 n.1062.

Age at first lambing and ewe longevity as mitigation options in sheep production

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Sheep production contributes 3% to global livestock greenhouse gas emissions (GHG), with demand for sheep meat projected to rise towards 2050. This implies a massive reduction in emission intensities (i.e., GHG emission from production of one unit of product, here sheep carcass). To mitigate emissions, we studied reduced age at first lambing and increased ewe longevity using the HolosNorSheep model. HolosNorSheep uses a cradle-to-farm gate system boundary and IPCC methodology to estimate GHG emissions from Norwegian sheep production systems. The model estimates yearly emissions of direct CH₄ from enteric fermentation and manure management, direct and indirect N₂O from manure management and soils, and CO₂ emissions from production and use of farm inputs. The calculations were performed keeping the number of winterfed ewes in the herd constant, i.e. total carcass production was allowed to increase above the baseline. Thus, when improving longevity (i.e., age at culling from two to seven years) or reducing the age at first lambing (i.e., from two to one year), the number of ewe lambs for replacement was reduced, thereby increasing the number of lambs for slaughter. Carcass yield was further influenced by the curvilinear relationships between ewe age and the number of weaned lambs, growth rate and carcass weight, and the number of culled ewes. The emission intensity decreased by 16% when ewes lambed as yearlings compared to at two years. Culling ewes at two years yielded the highest emission intensity, decreasing with age and reaching a minimum at five years (-5% compared to culling at two years). Mating ewe lambs and optimizing ewe longevity appear to be effective mitigation strategies in sheep production.

Slaughter results and evaluation of carcass quality of meat-type breed lambs in Latvia

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A study conducted from 2020 to 2023 in Latvia, at the "Klimpas" ram testing station of the Latvian Sheep Breeders Association. Feed control was conducted on 99 lambs: Ile-de-France (36), Charollais (21), Texel (21) and Dorper (21) breeds. Lambs were fed unrestrictedly with commercially produced compound feed and grass hay prepared at the station. Lambs were slaughtered in a certified slaughterhouse, preceded by a 12-hour fasting period. After slaughter, carcasses were weighed, measured, and evaluated for quality according to the EUROP (respectively 1 to 5 points) classification system. Results were analysed in software "R" using unpaired t-test, $p < 0.05$. The study found that significantly younger slaughtered lambs of the Ile-de-France breed were 130.5 ± 1.83 days old with an average live weight of 47.30 ± 0.45 kg. Significantly older lambs were of the Dorper breed, 147.0 ± 2.16 days, a difference of 16.5 days ($p < 0.05$), with a pre-slaughter average live weight of 46.49 ± 0.51 kg. The highest carcass yield was obtained from Charollais and Dorper breed rams, at $47.7 \pm 0.43\%$ and $47.5 \pm 0.40\%$, respectively. The best muscle grading was for Texel (1.9 ± 0.07 points) and Charollais (2.0 ± 0.14 points) breed lambs, while the least fatty were Charollais breed lamb carcasses, averaging 2.2 ± 0.09 points. Quality assessment of Ile-de-France and Dorper breed lamb carcasses was equal, with muscle development at 2.4 ± 0.09 points, but fatness at 2.8 ± 0.05 points and 2.8 ± 0.09 points, respectively. Acknowledgements. The study was funded by the LZP-2021/1-0489 project.

Essential oils effects in in vitro sheep proliferation and cytokine secretion

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In the last decade, the effects of essential oils (EOs) were deeply investigated for their role as rumen modifier, antioxidant and anti-inflammatory on livestock. The objective of this experiment was to study the action of *Mentha x piperita* L. (M), *Rosmarinus officinalis* L. (R), and *Lavandula angustifolia* L. (L) EO on in vitro sheep model of inflammation. Peripheral blood mononuclear cells (PBMCs) were isolated from sheep blood by density gradient, and treated with four concentrations (3, 5, 7, and 10%) of each EOs. PBMC were stimulated with Concanavalin A (ConA) and the combination of ConA and lipopolysaccharide (LPS), to test viability (XTT assay), proliferation (Bromodeoxyuridine assay), and interleukin (IL)-6, IL-10, and IL-8 (ELISA) production after EOs treatment. Data were analyzed by using one-way analysis of variance with Tukey post-hoc adjustment for multiple comparisons. The viability was not affected by EOs treatments. The BrDU incorporated during proliferation increased significantly when MEO was added at 10% and in presence of ConA and LPS. Moreover, REO treatment at 5% registered higher proliferation than at 7 and 10% with ConA and LPS. Finally, cells treated with 10% of LEO in presence of ConA and LPS resulted in the highest proliferation except for that at 10% stimulated with ConA. Cytokine profile was influenced in relation to the type of EOs in vitro treatment. Present results encourage further in vivo studies by using EOs as feed additives especially to support immune competence in livestock.

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Poster 27

Feeding management in Spanish Assaf dairy sheep farms

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Feed management is one of the determining factors in the sustainability of dairy sheep farms. In order to study the relationship between production and feeding management, 20 commercial Assaf dairy sheep farms located in Castilla y León (Spain) were visited. In each farm, information related to the conditions of the facilities intended for dairy ewes was collected. Milk production (l/ewe/year) of 60% and 40% of farms was less or greater than 560 l, with average values of 444 and 642 l respectively. Data was analysed by GLM ANOVA procedures, using IBM SPSS V 29. The size of the flock (1338 ewes/farm), the density (1.6 m²/ewe) and feed consumption (3.0 kg of DM/ewe) did not present significant differences ($P > 0.05$) among production groups. Farms with more than 560 l of production showed greater accessibility to feeders ($P < 0.05$, 0.45 vs. 0.34 m/ewe) and joint score (space, cleanliness plus accessibility) of the feeders ($P < 0.01$, 3.1 vs. 2.0 points). The number of feeding lots, type of ration, administration frequency and place to administer feed in each farm were not statistically different ($P > 0.05$) among the production groups. Sixty-five per cent of farms made two feeding batches: high and medium-low production, 5% a single batch and 30% of the farms differentiated 3 or more feeding batches. Eighty percent of the farms used feed as a total mixed ration, with an administration frequency of once or twice a day in 70% and 25% of the farms respectively. The most frequent place to administer feed was troughs (45%), aisle (35%) and troughs with belts in 20% of the farms. Our results showed that the mechanization and management of feeding has been facilitated by the high production of Spanish Assaf dairy sheep. Grant PID2020-113395RB C22 funded by MCIN/AEI/10.13039/501100011033.

Methionine balancing diet during transition and early lactation periods increased milk yield and composition in dairy ewes improving commercial farm profitability

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Recent research reported dietary recommendations of digestible methionine (MetDi) in dairy ewes similar to those of dairy cows. Objective was to confirm, at commercial farm level, the effect of MetDi supply on milk yield and composition in dairy ewes. Assaf ewes (525) in a commercial farm, Valladolid, Spain were fed for 3 weeks before lambing to 3 months after. A TMR diet was fed ad libitum formulated to meet INRA2007 nutrient requirements. Treatments were two diets (18.1% CP, NEL=5.96 Kcal/kgDM): basal diet “CTR” and MS plus methionine supplementation (MetaSmart Dry; isopropyl ester of 2-hydroxy-4-methylthio butanoic acid, Adisseo) provided 71 and 100% of MetDi requirement, respectively. MetaSmart Dry was administered in the TMR with 9 and 12g/animal/day pre- and post-partum periods. The trial was conducted as a 2-groups randomized block design. Ewes were randomly allocated to experimental treatments. Individual milk yields (MY) were recorded daily and milk fat and protein monthly. Data were analysed using the MIXED procedure of JMP for repeated data with treatments as fixed factors. MY is higher in ewes fed MS before lambing (2.97 vs 2.80 kg/d, prepartum: P<0.05), milk protein content did not differ. Ewes fed MS decreased milk fat content (6.01 vs 5.63 %, lactation: P<0.01). Milk protein yield increased in ewes fed MS before lambing (132.4 vs 127.5 g/d, lactation: P<0.01). Ewes fed MS before lambing had higher milk fat yield (149.5 vs 142.9 g/d, prepartum: P<0.01). In Summary, balancing diet for methionine during transition period before and after lambing improved performances of lactating dairy ewes.

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Theatre 1

Host genes shape the lower gut microbiota of Holstein cows: evidence from sequence-based GWAS

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In Holstein cows, our previous works have highlighted the influence of host genetics on the lower gut microbiota, with significant heritability estimates of taxa abundances ranging between 0.05 and 0.22. Additionally, genetic correlations between the lower gut taxa and traits of interest (production, health, fertility) have been observed. In this study, we aim to explore the genetic architecture of the composition of the lower gut microbiota. To this end, samples of 1,875 Holstein cows from 144 French commercial farms were collected and sequenced using 16S rRNA gene sequencing. Cows were genotyped with the 50k SNP array and the genotypes were imputed to the sequence-level using the ninth run of the 1,000 bull genomes project reference panel. The centered-log ratio abundances of 40 heritable genera were then adjusted for non-genetic effects. Genome-Wide Association Studies (GWAS), conducted on approximately 13 million variants, identified seven Quantitative Trait Loci (-log(p-value)>7.3) and gene-based analyses revealed 37 underlying candidate genes. Notably, Turicibacter abundance was found to be associated with CMCI1, an immunity-related gene expressed in the gastrointestinal tract of cattle. We also highlighted a significant association between the abundance of Akkermansia and ABO, a gene known to impact the gut microbiota composition in humans and pigs. To summarize, this study identifies specific regions of the bovine genome that influence the composition of the lower intestinal microbiota, thereby advancing our understanding of the holobiont concept.

Decoding the Genetic Dialogue: Rumen Microbiome and Host Genome Interaction in Enteric Methane Emission

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This study encompasses a comprehensive analysis of 493 cows rumen and blood samples from one commercial Holstein farm in Denmark, integrating 16s rRNA and metagenome sequencing, alongside blood transcriptome on the rumen sampling day. Rumen and blood samples underwent NMR analysis. Cows were genotyped using a 50k SNP array. Daily feed intake was recorded via 3D camera (CFIT), and methane emissions were taken using a sniffer method. Extensive production, feeding, and management data are available, prompting the exploration of multiple research questions – 1) investigating the microbial compositions within the rumen to enhance our understanding of their fundamental role, 2) assessing whether metagenome data provides a more robust microbiome profile compared to 16s rRNA, 3) exploring the relationships between metabolites present in the rumen and blood samples, 4) investigating whether the rumen microbiome has an impact on the blood transcriptome profile, 5) scrutinizing the interplay between the rumen microbiome and the host genome concerning enteric methane emission, 6) examining the correlation between daily feed intake and daily methane emission, shedding light on potential influencing factors, 7) assessing whether the integration of omics data with genotypes improves the reliability of genomic prediction. Furthermore, the study aims GWAS with imputed whole genome sequence variants level focusing on enteric methane, feed intake, targeted metabolites in rumen and blood, and microbiota abundances. The outcomes are expected to contribute to a deeper understanding of the role and interactions of the rumen microbiome with host genetics and in adaptation of breeding strategies to mitigate enteric methane emission from dairy cows.

Impact of microbiome on genomic predictions for methane emissions in Holstein cows

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The rumen microbial composition is partly determined by host genetics, and its variation has been associated with variation in methane emission, offering a potential tool to mitigate indirectly greenhouse gasses via animal breeding. So far, breeding approaches that include microbial information are hindered by low sample sizes. In this study, we collected rumen fluid samples from 955 Holstein cows from 12 commercial farms in the Netherlands and analysed them via long-read metagenomic sequencing. To the best of our knowledge, this is the biggest dataset of rumen microbial samples collected in one country worldwide. Individual daily methane concentration, pedigree, genotypes, and milk records for the cows are also available. This study aims to investigate the impact of microbial composition on genomic predictions for methane emissions. Thus, we will use a previously developed pipeline and implement three genomic prediction scenarios for methane emissions: 1) a model using only genomic data (benchmark); 2) a model using only microbial data (microbiability); 3) a model jointly using both genomic and microbial data. Each scenario will be validated to estimate the accuracy and bias of genomic predictions. This study is part of the Re-Livestock EU Horizon project and the Knowledge and Innovation Agenda of the Dutch Ministry of Agriculture, Nature, and Food Quality.

Why should we care about the rumen microbiome in next animal breeding strategies

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In the past decade, the exploration of microbial communities within animals has emerged as a focal point in studying animal genetics. Animals harbor a secondary genome comprised of microorganisms collectively referred to as the "microbiome." The rumen microbial community assumes a pivotal role in processes such as feed digestion, feed efficiency, methane emission, and overall health. The rumen microbiome and the host animal operate in symbiosis impacting the phenotype. Our previous studies showed that the rumen microbiome composition can be aggregated in few variables or principal components that explain more 25% of microbial composition and have moderate heritabilities. In this study we used over 500 Holstein cows from a Spanish reference population, examining their contributions to feed efficiency, methane emissions, and the underlying genetic control exerted by the cow over these communities. Employing Principal Component Analysis (PCA) and Partial Least Squares (PLS) analysis, we aggregated microbial composition into a few variables. These variables showed heritabilities ranging from 0.20 to 0.40, with large and positive genetic correlation, and are correlated with feed efficiency and rumen dysbiosis. Biological interpretations of these variables are shown, showing the importance of a proper balance between the cow genome and her metagenome. Thus, breeding goals should incorporate rumen microbiome health before encountering negative correlated effects from selection on improved feed efficiency and reduced methane production. Beyond microbial composition, our investigation extended to the presence of microbes from oral samples that are potentially associated with dry matter intake, and can be used as a proxy of feed efficiency.

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Theatre 5

Cattle genetics influences the abundance of ruminal pathogenic microbes

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Ruminal health refers to the capacity to effectively control and resist pathogenic microbes and could serve as an indicator of the immune system's efficacy in preventing their proliferation. Thus, it could be used as a resistance trait in breeding programs. Recently, microbiome-driven breeding has been developed to mitigate methane emissions and improve performance traits based solely on the composition of the ruminal microbiome. The aim of this research was to investigate whether this approach could be extended to enhance ruminal health. Host genomic (genotyped using 35K array and imputed up to 70K) and ruminal metagenomic (whole shotgun metagenomic sequences of the ruminal microbial DNA) data were available from 1,443 crossbreds between British Blue x Holstein, reared at performance test stations. From the metagenomic sequences, the abundances of 1,161 microbial genera were generated and centered log-ratio transformed before genomic analyses using Bayesian statistics. We identified genera containing well-known opportunistic pathogenic microbes with relative abundances ranging from 0.02% to 2.87%. The heritabilities of these microbes ranged from 0.17 (HPD95% 0.11 to 0.25) for *Staphylococcus* to 0.28 (HPD95% 0.19 to 0.39) for *Escherichia*. *Campylobacter* exhibited the highest negative genetic correlation with average daily gain at -0.26 (HPD95% -0.54 to 0.02). These findings suggest that microbiome-driven breeding (including potentially beneficial microbes) could be utilized to reduce the prevalence of opportunistic pathogenic microbes and improve animal health.

Large scale analysis of the cattle rumen microbiome in relation to methane emissions

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Methane emissions of dairy cattle are associated with the rumen microbiome. To predict or mitigate cattle methane emissions, detailed knowledge of the rumen microbiome is of advantage. So far, few studies have investigated methane emissions and its relationship to the rumen microbiome using modern long-read metagenomic (LRM) methodology. In this study the microbiome of 955 Dutch Holstein dairy cows from 12 different farms were sequenced using LRM while recording methane emissions with sniffers in automated milking systems. Also related metadata (genotype, diet, milk records, etc.) was gathered. The LRM data was analysed using SqueezeMeta software and >3500 species of microbes were found across all animals. This study aims to use the dataset to provide new insights into the rumen microbial abundance and diversity to a great level of detail. Moreover, to analyse the heritability of species, and to identify which species are associated with methane emissions. This will identify microbial species relevant for breeding for reduced methane emission. This study is part of the Re-Livestock EU Horizon project and the Knowledge and Innovation Agenda of the Dutch Ministry of Agriculture, Nature, and Food Quality.

Host genetics and hindgut microbiome jointly associate with daily milk yield in Holstein cows

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The microbiota in the gastrointestinal tract, together with host genetics, are critical for maintaining the health status and production performance of dairy cattle. Although the hindgut microbiome is feasible for sampling at a large scale, the interaction between the hindgut microbiome and host genetics and the potential association of the hindgut microbiome with daily milk yield (DMY) are less characterized. Therefore, this study aimed to estimate the heritability of hindgut microbiota in dairy cattle, calculate the microbiability (m_2 , defined as the proportion of phenotypic variance explained by microbiota) and identify the microbial marker for DMY. In this study, the profile of hindgut microbiota by 16s rRNA gene sequencing and host genetics by 150k bovine SNP genotyping of 646 lactating Holstein cows with DMY records were available for analysis. The heritability of 1862 microbiota traits including 24 bacterial community traits and 1838 single-taxon traits were estimated. The results showed that 75% of microbiota community traits and 24% of single-taxon traits had significantly non-zero h_2 ($P < 0.01$), and the estimates of h_2 varied from 0.08 to 0.54. The m_2 of DMY was 0.29, which is close to its heritability ($h_2, 0.31$). Both m_2 and h_2 of DMY decreased when considering both random microbial and genetic effects in the BLUP model. A total of 140 taxa were identified as microbial markers of DMY of which 32% had significantly non-zero heritability estimates. This study illustrated the influence of host genetics on the hindgut microbiota and the association of the hindgut microbiota with DMY. The results of this study might help provide innovative dairy cattle breeding programs using microbiome information.

Investigation of the Interrelationship between Ruminal and Oral Microbiomes of Angus Bulls

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The gut microbiome of ruminant animals plays an essential role in their digestion. Therefore, methodologies that facilitate such investigations are valuable. The current study investigated the interrelationship between the ruminal and the oral microbiomes of Angus bulls with the intention of establishing their level of relatedness. A total of 541 bulls from five different farms, located in three different states of the United States were used in this study. Ruminal contents were collected by esophageal tubing, while samples from the oral cavity were obtained by rubbing sterile swabs inside the mouth. Samples were immediately frozen in liquid nitrogen until examination of their microbial composition by analysis of the 16S rRNA gene was performed. Overall, the ruminal and the oral samples had very distinct microbial profiles. Ruminal samples had greater number ($P < 0.001$) of amplicon sequence variants (ASV), greater microbial diversity (expressed as Shannon diversity index; $P < 0.001$), and greater microbial evenness ($P < 0.001$). Beta-diversity (unweighted UniFrac distance) was also significantly different ($P < 0.001$) between the ruminal and oral samples. A total of 4323 microbial species were observed in the rumen and oral cavity combined, but only one third (33.1%) of those species occurred in both the rumen and oral compartments. Moreover, the relative abundance of only 30.7% of the species were found to be significantly different between the rumen and the mouth of the bulls. Taken together, these results indicate that the ruminal and oral environments are substantially different, and that predicting the ruminal microbiome based on the oral microbiome may not be a viable strategy.

Genetic selection for pig faecal enterotypes affects host upper gut microbiota and immune traits

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The faecal microbiota of 60-day-old Large White pigs reared in same conditions can be structured into two enterotypes, for which the keystone genera are Prevotella (P) and Mitsuokella (M) or Ruminococcus (R) and Treponema (T). We generated two pig lines (HPM and HRT) selected for a higher relative abundance of either P and M or R and T, respectively. Each line showed an increase in the prevalence of the selected enterotype over three successive generations. We used 40 animals per line from the 3rd generation to investigate if at 60 days of age they display differences in the microbiota of the small and large intestine, and in immune traits. Using 16S gene sequencing, the two lines were shown to harbour strong microbiota differences at the descending colon (648 out of 1334 ASV and 79 out of 144 genera found differentially abundant), with less differences at the ileal Peyer's patches (38/435 ASV and 18/126 genera). Piglets from the HPM line exhibited a higher number of eosinophils and natural IgM, while piglets from the HRT line showed higher counts of CD4⁺ CD8⁺ T cells and stronger phagocytosis activity. Overall, our results show that direct genetic selection for the composition of the faecal microbiota is not only associated with changes in the microbiota of other sections of the gut, but also with host traits related to immunity. The HPM and HRT divergent pig lines are therefore a powerful tool for better understanding the combined effects of host genetics and gut microbiota on a range of phenotypes relevant to sustainable livestock systems, including health and immune traits.

Exploring the role of microbiota on phenotype expression under different biological assumptions

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Here, we explored the interaction of microbiota (M) with genome (G) and environmental effects (ENV) in influencing phenotypes (y) across six assumptions: m1: G and ENV jointly controlled y. m2: M and ENV collectively influenced y. m3: ENV, G, and M independently controlled y. m4: ENV, G, and M collectively regulated y, with M regulated by both G and ENV. m5: Similar to m4 but without a direct effect of G on y. m6: M explained only what G and ENV could not in y, i.e., residuals of m1 are regulated by M. Our dataset included 1124 individuals with both M and G data and measured for phenotypic traits: Average Daily Gain (ADG), Backfat (BF), and Carcass Weight (CW). When analyzing y variance explained by G, M, and ENV across models, G contributed 20%, 45%, and 22% of h² for ADG, BF, and CW, while M accounted for 60%, 49%, and 62% of the total variation. Combining G and M decreased h² compared to G alone; e.g., ADG went from 20% to 14%. This reduction was explained in m4, where 14% of h² was due to the direct effect of G on y and 8% due to the mediated effect of G by M. Due to consistent predictability across phenotypes, we focused on ADG when comparing r across different models. m1 had the lowest r (0.25), followed by m2 (r=0.32) and m5 (r=0.32). m6 had an r of 0.37. m3 and m4 had r=0.41. M models had better r compared to G alone. Considering G and M jointly enhanced y prediction, while decomposing the effect of M in m4 did not significantly improve predictions. However, when comparing genetic effects obtained from the whole and partial databases, m4 had the highest correlation (r=0.60), followed by m3 (r=0.56).

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Theatre 11

EU-CIRCLES project: Evolution of pig microbiota and health under different farming conditions

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This study aimed to track the evolution of the pig's microbiome from early colonization to slaughter, focusing on identifying microbial species and functions associated with the health status of pigs raised in different rearing conditions. Ninety-six piglets, from 22 litters, were divided into two groups — PC1 (Production chain 1) and PC2 (Production chain 2) — at 21 days of age and monitored until slaughter, pigs were fed the same diet through the whole trial. Feces and blood were collected at day (d) 21 (before weaning, T1), d42 and d80 (weaning unit, T2 and T3), d98 and d278 (fattening unit, T4 and T5), with slaughter occurring at d297 (T6). Shotgun microbiome analysis, blood formula, oxidative stress and immunoglobulin levels were examined. Rearing condition significantly affected species-level microbiome and functional profile at T2, T3, T4, and T5. At T3, piglets in the PC1 had a lower species alpha diversity (P = 0.002) and a higher abundance of *Prevotella* sp. P5-92 (LDA=4.0, P.adj<0.01), *L. amylovorus* (LDA=5.1, P.adj<0.01), and *B. porcum* (LDA=4.8, P.adj<0.01) compared to those in the PC2, which were characterized by *L. jhonsoni* (LDA=4.9, P.adj<0.01) and *L. reuteri* (LDA=4.7, P.adj<0.01). Additionally, at T3 microbial pathways associated with drug and xenobiotic metabolism were more active in pigs in PC1 (log₂FC= 1.5, p.adj= 0.04) that also showed a higher blood Neutrophil/Lymphocyte ratio (P<0.001). These findings underscore the significant impact of farm-specific conditions on pig microbiome development and health status.

Exploring mortality and culling causes of breeding does through gut microbiome analysis

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Mortality and culling are major concerns in rabbit farming due to their impact on production and economic losses. This study investigates the relationship between gut microbiota composition and mortality/culling cause in female rabbits. The study included 44 females does with recorded causes of culling and mortality. The females were divided into 3 groups; Culled for reproductive reasons (CR), Culled for health reasons (CH) and Deceased (D). Soft fecal samples were collected from all does after the last parity before death or culling and subjected to DNA extraction and 16S rRNA sequencing. After bioinformatic analyses the core microbiome of each culling/mortality group was identified and Partial least square discriminant analyses (PLS-DA) was used to assess its association with gut microbiome. The results revealed distinct core microbiomes among the culling and mortality groups. Group D had 22% of its core microbiome not shared with the other groups, followed by CR with 16%, and CH with 13%. Moreover, the PLS-DA final model discriminated between the three groups and the prediction accuracies were: 96% for D, 84% for CR group, and 88% for CH. The group D (Deceased) exhibited the most unique core microbiome and the highest prediction accuracy in discriminant analyses. We hypothesize that alterations in gut microbiota composition, potentially leading to dysbiosis, may contribute to the distinct classification of animals into group D. The initial findings of this study highlight the importance of understanding the role of gut microbiota in breeding animal culling and mortality. Knowledge of microbiota or specific taxa within it, that are associated with adverse outcomes, can contribute to improved animal health, productivity, and welfare.

Rumen microbiome, methane emission and blood analytes in grain-fed bison

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Rumen microbiome has a pivotal role on feed efficiency, greenhouse gas emission, and carcass quality of live-stock. Our aim was to investigate the associations of these with carcass quality in grain-fed bison. Blood and rumen content samples and carcass traits of 37 bull calves were collected at slaughter. Rumen microbial diversity was investigated via quantitative PCR, and the volatile fatty acid concentration was determined and used for methane emission. Complete blood count was determined. Animals were separated into groups according to the carcass grade: A1 (n=4, Hot Weight (HD)=260±46 kg, Yield percentage (YD)=56±1.6), A2 (n=11, HW=284±42 kg, YD=57±1.8), A3 (n=13, HW=289±30 kg, YD=57±1.7), and A4 (n=9, HW=300±30 kg, YD=58±1.5). HW and YD did not differ among groups. The number of copies per gram sample of total bacteria, total fungi, total protozoa, and total archaea ranged from 9.9×10^{11} to 1.1×10^{12} , 3.5×10^5 to 2.0×10^6 , 6.4×10^3 to 1.3×10^4 , and 4.8×10^9 to 7.0×10^9 . Group A1 was different from group A4 in terms of acetic acid, propionic acid, iso-butyric acid, butyric acid, iso-valeric acid, and valeric acid. In addition, methane emissions differed between the high-quality groups (A1, 4.5 ± 1.7 ; A2, 4.2 ± 1.5 mmol/L) and low-quality groups (A3, 2.5 ± 0.7 ; A4, 2.5 ± 0.7 mmol/L). The red blood cell count, haemoglobin, haematocrit, platelet, white blood cell counts were 9.5 to $9.7 \times 10^{12}/L$, 182 to 188 g/L, 0.49 to 0.52 L/L, 174.8 to $199.5 \times 10^9/L$, and 7.2 to $8.7 \times 10^9/L$. These did not differ between groups. Further analyses on blood biochemistry hormones are in due course. The determination of the genomic composition of the bison subspecies will be factored in this analysis. These preliminary results indicate individual variation and relationships between carcass grading with ruminal parameters to be further evaluated.

Fecal microbiota of pregnant mice in a divergent selection experiment

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The microbiota in animal production is increasingly recognised as a key factor influencing animal health, productivity and overall well-being. After implementing a selection experiment in mice, two divergent lines were obtained: a high and a low for birth weight environmental variability (H-line and L-line). The L-line was more robust when subjected to changes in temperature or feed restriction. The microbiota in pregnant mice provides a translational model for understanding similar processes in other prolific animals, the fecal microbiota composition was studied in these two mice lines to assess its association with robustness. Samples were used from 77 pregnant females from four generations of selection (43 for L-line and 34 for H-line). The 16S rRNA V4 region was sequenced, the amplicon sequence variants (ASVs) were determined using the DADA2 standard pipeline and the taxonomic assignment was performed using the SILVA v138.1 database. The alpha diversity was calculated and visualized in R using the phyloseq package and statistically evaluated by the Mann-Whitney U test. Beta diversity comparison was performed using the vegan and pairwiseAdonis2 packages in R. Non-parametric multivariate analysis of variance was used to assess the structure of the bacterial profiles using Bray-Curtis dissimilarity indices. Alpha diversity was not different between lines, suggesting that the diversity is equal within the sample regardless of the line, with a tendency for the L-line to have lower diversity maintained over generations. Regarding the beta diversity, the intra-sample variability was much lower in the H-line than in the L-line ($p < 0.001$). Microbiota differences may be associated with L-Line benefits.

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Poster 15

The lung-gut axis during polymicrobial respiratory infections in calves holobionts: the impact of microbiota dynamics on disease outcomes

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The gut-lung axis is a bidirectional communication system between the gastrointestinal and respiratory tracts that influences the host's health and disease susceptibility. However, the role of this axis on the calf holobiont defense against pathogens is poorly understood. In our study, we investigated dynamic changes in airways and gut microbiomes in 30 veal calves with varying disease severity from day 7 to 5 months of age, using 16S rRNA gene sequencing. We also sampled blood to evaluate the host's immune response. Notably, the bacterial diversity in the upper respiratory tract (URT) was lower than that of the gut and correlated with illness symptoms. Moreover, URT and gut microbiotas show different ecological dynamics, where the airway microbiome was more temporally variable. Yet, both respiratory and fecal microbiota reached synchronous homogenization from day 60 onwards. These findings shed light on previously unknown microbiome patterns and interactions between URT and gut microbiomes during respiratory infection diseases in calves. Further microbiota functional analysis and host immune response measurements are ongoing to understand the holobiont response and resistance to polymicrobial respiratory diseases.

Optimizing Piglet Growth and Meat Quality: The effect of Dietary Additives (KMAX, Solumarin) in Gut Microbial Balance

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Exploring the significant influence of dietary interventions on the gut microbiome in piglets is increasingly recognized for its potential to enhance livestock health and productivity. The gut microbiome is essential for nutrient absorption, immune function, and overall well-being. Our study aimed to investigate how different dietary treatments affect microbial diversity and composition. We utilized metagenomic sequencing to analyze the gut microbiome of piglets fed a standard diet with supplements KMAX or Solumarin. We collected stool samples from 25 piglets, extracting microbial DNA for analysis through Next-Generation Sequencing (NGS), specifically using the Illumina 16S V3-V4 amplicon sequencing approach. The analysis was conducted using QIIME2 and DADA2 bioinformatics tools. Our results showed that KMAX and Solumarin significantly influence the gut microbiome, notably altering the Firmicutes/Bacteroidetes ratio, an important gut health and metabolic function marker. This study underscores the potential of dietary interventions to improve livestock health, suggesting benefits for the intestinal environment, meat quality, and consumer health.

Rational Designed Microbial Consortia for Chicken: an in silico approach

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A good start is half the battle won, crucial for broiler chickens, and means providing optimal post-hatch conditions. This includes climate and early feeding tailored to various life stages. Gut health is a prerequisite for sustainable poultry production, reducing medication use. Previous research has shown that gut development starts immediately post-hatch. This includes microbiota colonization and morphological, functional, and immunological maturation. The last decade a plethora of studies have been performed in profiling the chicken gut microbiota, including the small intestine, caeca and cloaca/faeces. These studies have shown which microbial species reside in the chicken gut and which factors influence the microbiota composition. In the current system, stables are cleaned rigorously to kill potential pathogens, which unintentionally removes beneficial bacteria as well. Our aim is to design a chicken microbial consortia that increases the resilience in the immediately post-hatch period. We want to introduce a rational designed microbial consortium prior to microbiome maturation, thereby occupying the microbial niche that allows for pathogen colonization. To this end, based on available literature and data, we developed an in silico bioinformatic pipeline that aims to select 5-12 bacterial species that cover the core functions (~80%) of the chicken gut microbiota. Metagenomic sequencing data allowed identification of species and determination of their functional role. In this example, species were selected from a public database. However, ideally an in-house collection of known species isolates is used to ensure cultivation under laboratory conditions. Taken together, we have set-up a bioinformatic pipeline that identifies the microbial function utilizing metagenomic sequence data.

Dynamic Distribution of Gut Microbiota in Pigs at Different Growth Stages

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In the swine industry, understanding the relationship between intestinal microbiota and growth stage is essential because growth stage is directly related to the feeding system of pigs. 709 fecal samples were collected from 179 pigs at 4 time points, including the 1st day, the 21th day, the 50th, and the 100th day after the birth of swine, were used for microbiome analysis. For the microbiome analysis, the fecal microbial richness increased over time from day 1 to 100 and the β -diversity of fecal microbiota was separated significantly at different time points. One-way ANOVA and post hoc Tukey's HSD test for multiple mean comparisons were used to find significant differences in alpha diversity and microbial taxa between the growth stages. One-way ANOVA was performed using the R statistical package and significance was assumed at $P < 0.05$. A dynamic shift is confirmed in the gut microbiota of pigs at different ages and growth phases. Bacteroidaceae and Enterobacteriaceae are dominant in piglets before weaning, then Prevotellaceae shift to be the predominant at the family level. In conclusion, this study elucidated the dynamic changes of porcine fecal microbiota at different ages and growth phases, which may provide a reference for a comprehensive understanding of the intestinal health status of pigs at different growth stages and investigating host-microbe interactions in the swine industry.

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Poster 19

Unraveling the influence of the host genetic on vaginal microbiota composition in Holstein cows

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The vaginal microbiota is essential for maintaining reproductive health, bolstering immune function, and enhancing herd productivity in domestic animals. In this context, we have recently unveiled relevant associations between cow vaginal microbiota relative abundances and host traits linked to health and dairy production traits. However, the influence of host genetic makeup on the bovine vaginal microbiota, along with their genetic correlations with animal performance, remains mostly unknown. To address this question, we performed a comprehensive genetic study of the vaginal microbiota in Holstein cows. Vaginal swab samples were collected from 681 cows in 18 commercial herds from northern France between 2017 and 2018. Additionally, fertility, milk production, and health traits were recorded for each cow. Vaginal microbiota samples were sequenced for the 16S rRNA gene, and count tables for amplicon sequence variants (ASV) were generated. We estimated the genetic parameters associated with vaginal microbiota for α -diversity (Chao1, Shannon, Simpson) and β -diversity (Bray-Curtis dissimilarity) indices, as well as for the centered-log ratio (CLR) abundances of 64 genera, 48 families, 38 orders, and nine phyla reaching a 50% prevalence threshold. Our study will estimate heritabilities of the vaginal microbiota, as well as genetic correlations between taxa and host phenotypes. In future work, after imputing all the cows at the sequence level, we plan to perform Genome-Wide Association Studies (GWAS) to identify specific genes and genomic variants involved in the genetic determinism of the vaginal microbiota composition.

Study of holobiont resilience following symbiotic transmission disruption in two rabbit lines selected for feed efficiency

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This study was carried out on two lines, each selected for a feed efficiency criterion (growth on restricted feed for the AlimR line or residual consumption on ad libitum diet for the ConsR line), introduced in a building by adoption of newborn rabbits by SPF females. Following this break in symbiotic transmission, females feces samples was used to study the composition of the microbiota of the two lines over 7 generations (17 females per line and per generation). The proportion of Firmicutes was significantly higher in the AlimR line than in the ConsR line (82.22% vs 78.21% respectively, $p < 0.0001$). Conversely, the proportion of Bacteroidota was significantly higher in the ConsR line than in the AlimR line (20.38% vs 16.50% respectively, $p < 0.0001$). The effect of generation was also significant for Firmicutes and Bacteroidota abundance ($p = 0.0009$ and $p < 0.0001$, respectively). The Shannon diversity index was significantly higher in the AlimR line than in the ConsR line, (5.23 vs. 5.14 respectively, $p = 0.02$). The effect of generation was also significant for the Shannon index ($p = 0.003$). Finally, a discriminant principal component analysis of the composition of the microbiota allowed to distinguish between the two lines and between generations, in particular the 11th generation of selection corresponding to animals adopted by SPF females and the 18th generation. In conclusion, after a break in symbiotic transmission following the adoption of young rabbits by SPF females, we demonstrated differences in taxonomic profiles, diversity and composition of the fecal microbiota between two lines selected for different feed efficiency criteria and between generations.

Both age and genetic predisposition to mastitis influence the digestive microbiota in ewes

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The microbiome is known to influence the immune system. In humans, age-related dysbiosis and inflammation have been linked, but little is known in livestock. The aim of this study was to evaluate the digestive microbiota and its associations with age and genetic background for mastitis. Forty Lacaune dairy ewes were included in the study, 20 ewes with ≥ 5 lactations (old) and 20 ewes in first lactation (young). They were equally divided into two divergent lines selected for mammary inflammatory response based on milk somatic cell count (SCC-, resistant and SCC+, susceptible to mastitis). Ewes were managed as a single group for several months at the end of the lactation. After euthanasia, digestive content was sampled at different sites, and bacterial DNA was sequenced using the 16S rRNA gene. FROGS pipeline and mixOmics package were used for bioinformatics and statistical analysis. Based on the ruminal microbiota, "young" ewes can be distinguished from "old" ewes using an sPLS-DA. Furthermore, within the "young" group, SCC- and SCC+ can be distinguished, whereas in the "old" subgroup, SCC- and SCC+ cannot be discriminated according to genetic line. In contrast, when the analysis was applied to faecal content, four distinct groups were identified. This study shows that the digestive microbiota of ewes varies with age and genetic background for disease susceptibility. Faeces might be a better matrix than rumen to discriminate microbiota changes about the predisposition to inflammatory responses.

Ruminant microbiome data are skewed and unFAIR, undermining their usefulness for improving sustainable production

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Ruminant microbiome composition varies depending on factors such as ruminant species, habitat, and diet. This underscores the importance of having a good representation of ruminant microbiomes in their local environment to translate research findings into practical applications. Unfortunately, this information is currently lacking. In this study, we delved into the metadata of microbiome studies involving farmed ruminants to assess global representativeness and categorize information based on ruminant species, geographic location, body site, and host information. Utilizing data from the International Nucleotide Sequence Database Collaboration database, we analyzed 47,628 sample metadata, with cattle accounting for over two-thirds of the samples. In contrast, goats with a worldwide population similar to cattle were markedly underrepresented, comprising less than 4% of the total samples. Most samples originated in Western Europe, North America, Australasia and China but countries with large ruminant populations in South America, Africa, Asia, and Eastern Europe were underrepresented. Microbiomes from the gastrointestinal tract were the most frequently studied, constituting about 87% of all samples. However, there has been a noticeable increase in samples from other body sites such as the respiratory tract, milk, skin, reproductive tract, and fetal tissue over the past decade. More than 40% of the samples lacked basic information, and many were retrieved from generic taxonomic classifications, requiring manual recovery and curation. The absence of data on diet, production system, age, or breed limits the data reusability for further analysis and follow-up studies. Taxonomic assignment of the ruminant host and a minimum set of metadata attributes using accepted ontologies tailored to host-associated microbiomes are prerequisites for this. Public repositories should mandate the inclusion of this information to enhance data reusability. The results from this survey highlight the need to encourage studies of the ruminant microbiome from underrepresented ruminant species and underrepresented countries worldwide.

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Poster 23

Genetic-Microbiota Interplay in Laying Hens: Enhancing Vaccine Response and Reducing Antibiotic Reliance in Poultry Production

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Vaccination stands as the primary and most effective approach to prevent infectious diseases in livestock; however, the efficacy of vaccines often varies. Understanding the factors influencing efficacy, including genetic variations in the host and gut microbiota composition, is crucial. In a first experiment, we compared four groups (50 per group) of two chicken lines (Rhode Island Red (RIR) and Leghorn (LEG)), assessing the impact of a three-antibiotics cocktail and rearing conditions (fully indoor or with outdoor access) on vaccine response. This revealed antibiotic-induced changes in the gut microbiota for both lines, with changes affected by the interplay between antibiotics and housing conditions. Notably, the LEG line showed reduced vaccine responsiveness across various vaccines, prompting a deeper exploration into underlying genetic and microbial factors. Thereafter, in a second experiment focus shifted to 400 individual LEG hens, using Genome-Wide Association Analysis (GWAS) to identify genomic regions associated with both vaccine response and microbiota composition. Preliminary findings indicated distinct genetic control of both vaccine responsiveness and gut microbiota composition in the LEG line, paving the way for deeper exploration. Our approach not only enhances vaccine response but also addresses overall health and production outcomes, with implications for selecting hens with improved vaccine response, enhanced health, and reduced antibiotic reliance in sustainable poultry breeding practices.

Can semen microbiome contribute to improved prediction of bulls' fertility?

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Reproductive performance complexities, including sperm quality and fertility, are influenced by multifactorial processes, with seminal microbiome emerging as a crucial factor impacting fertilization dynamics and pregnancy outcomes. Paving the way for advancements in the emerging field of hologenomics, integrating seminal microbiome data into genomic prediction models holds promise for accelerating fertility improvement in farm animals. Leveraging the Angus Australia database with data from 78,555 animals, we simulated traits such as percentage of normal sperm (PNS) and prolificacy based on progeny DNA pooling for 713 sires. Real semen microbiome data from 45 bulls were used as a template for the simulation of semen microbial profiles for the 713 sires. Our integrated models, combining genomic and microbiome data, explained a larger proportion of phenotypic variation for PNS (0.94) and prolificacy (0.56) compared to single-source models (e.g., 0.36 and 0.41, respectively, using only genomic information). Additionally, the integrated model resulted in greater phenotypic differences between bulls ranked in the top and bottom quartiles of predictions, suggesting the potential for enhanced productivity and sustainability in livestock farming. Our findings highlight the semen microbiome's role in improving cattle fertility traits, paving the way for innovative selection strategies and interventions to enhance reproductive performance in livestock.

Spray dried plasma protein as functional protein source enhanced gut health in post-weaned pigs

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The present study explores the impact of dietary SDPP (196 g/kg) on microbial and molecular “signatures” involved in resilience and gut health of growing pigs (average initial body weight of 34.9±3.4 kg), using a comprehensive FeedOmics approach. The latter uses a series of ~omics-based techniques including microbiome, transcriptome, and metabolome analyses. A diet based on soyabean meal (SBM) as single protein source in the diet was used as reference. Compared to ileum, jejunum emerged as a focal point of the effects of SDPP and exhibited a dynamic bacterial ecosystem with significantly ($P < 0.05$) reduced alpha diversity indices compared to the reference diet. We noted a significant ($P < 0.05$) decrease in the relative abundance of *Sarcina* genera in the small intestine. This reduction mirrors the supportive effect on gut health observed with pharmaceutical concentrations of zinc oxide and antibiotics. Transcriptomic analysis identified 319 differentially expressed genes ($FDR < 0.05$, $|\text{Log}_2\text{FC}| > 1$ or < -1) in jejunal tissue. These genes revealed pathways linked to maintaining intestinal homeostasis and locally reducing immune system activity. The up-regulation of tight junction receptors, particularly in the jejunum, suggested an enhanced effect on intestinal integrity. Moreover, the identification of significant metabolites in blood of SDPP pigs, such as threonine, taurine, and glutamine/glutamate. These metabolites are vital for regulating cellular energy, coordinating immune responses, and aiding in the restoration of tight junctions within intestinal tissue, thereby making a positive contribution to gut health. Overall, the results provide valuable information on understanding the intricate interactions at the microbial and molecular level that highlights the functional value of SDPP in growing pigs.

Reduced α -diversity of bacteria in refrigerated compared to non-refrigerated colostrum from primi and multiparous dairy cows

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The ingestion of microbes present in colostrum by calves is thought to influence development of the immune system and gastrointestinal tract. The colostrum microbiome (colostrone) has been associated with passive immune status of dairy calves. The study objective was to determine effect of refrigeration and reheating on the composition of the colostrum microbiota. Twenty-five Holstein- \times and 15 Jersey cows were milked within 2-6h of parturition. Thirteen colostrum samples were refrigerated for 24h (4°C) and reheated (38°C) prior to feeding. Before feeding, 10 mL aliquots of non-refrigerated (DD) and refrigerated (RH) colostrum were collected, snap-frozen in liquid nitrogen and stored at -80°C. Illumina amplicon sequencing of the V4 region of the 16S rRNA gene was performed on extracted DNA. Data underwent PERMANOVA, α - and β -diversity analyses using R packages DADA2, Phyloseq, Microbiome and Vegan. Taxonomy was assigned using the SILVA database (v. 138.1). There was no effect of parity or breed. Reduced α -diversity ($P=0.0001$) was observed in RH compared to DD colostrum. The bacterial phyla identified with the highest relative abundances were Bacillota (42.5%), Pseudomonadota (34.2%), Actinomycetota (11.2%), and Bacteroidota (7.6%). Several pathogens were identified including *Yersinia* and *Klebsiella*. The archaeal genus with the highest relative abundance was *Methanobrevibacter* (85.5%). This is the first investigation on effect of refrigeration on the colostrone. Refrigeration reduces the α -diversity of the colostrone and may affect the development of the calf's GI microbiome.

DonKeyWorld: a comprehensive study on *Equus asinus* from biodiversity management to milk nutraceutical and technological properties

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The most promising use of donkey is towards milk production thanks to recent findings on its nutraceutical properties. Thus, the overarching goal of the DonKeyWorld project is to generate fundamental knowledge on the Italian donkey sector. Specifically, one of the three main objectives is to further validate the nutraceutical effect of donkey milk via the characterization of foal microbiota. Thanks to the presence of foals with jennies throughout lactation, the composition of the microbiome in the foals' gastrointestinal tract can be seen as a reflection of milk quality. The presence and abundance of bacterial taxa and diversity measures can be used as indicators of foal gut health status. Combining a comprehensive characterization of jennies' milk with foals' microbiota characterization (via 16S sequencing), we will identify milk quality parameters that can be beneficial or antagonistic towards foal's health. We expect to collect microbiological samples at two time points from at least 80 foal-jenny pairs belonging to the Romagnolo breed and being raised in a large single farm. At thirty days in lactation, an individual milk sample from each jenny will be collected and analyzed for its nutraceutical and technological properties. This research was supported by the project "DonKeyWorld", funded by the Italian Ministry for Universities and Research [project code P20228Z3RJ (PRIN)]

Relationship between performance phenotypes and rumen microbiome and liver transcriptome data of beef cattle using a systems biology approach

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Provision of feed in beef production systems is a major determinant of profitability, thus identifying and breeding cattle with improved feed efficiency will benefit the beef sector. The objective of this study was to examine the relationship between animal performance phenotypes including residual feed intake (RFI), dry mater intake (DMI) and average daily gain (ADG) with both rumen microbiome and liver transcriptome data using a network-based systems biology approach. In the network, DMI had the greatest number of microbial and gene connections ($P < 0.05$) followed by RFI and ADG. Hepatic genes related to lipid metabolism were connected to both RFI and DMI, whilst genes related to immune response were connected to DMI. Microbes were not commonly directly related to any of the phenotypes examined, with the exception of the *Succiniclasticum* genera, which was negatively connected to both RFI and ADG. Results from this study highlight the complex interplay between rumen microbiome and hepatic transcriptome data with key economically important traits in beef cattle and enhance our understanding of the biology underlying RFI, DMI and ADG. Acknowledgement: This research was funded by the Irish Department of Agriculture, Food and the Marine (RSF13/S/519). Kate Keogh received funding from the Research Leaders 2025 programme (co-funded by Teagasc and the European Union's Horizon 2020, Marie Skłodowska-Curie grant agreement number 754380).

Session 56

Theatre 1

Automated real-time activity monitoring in infectious disease experiments with various livestock species

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Automated real-time monitoring of animal activity can improve the quality of animal experiments. It provides insights into a treatment's effect and helps to refine experiments, e.g. through better detection of humane endpoints. This study aimed to develop a tool for automated real-time monitoring of animal activity in infectious disease experiments with various livestock species in group-housing and under high containment conditions. We explored various technologies and concluded that the vectorial dynamic body acceleration (VDDBA) derived from accelerometers is currently most promising to obtain robust and informative time series. We demonstrated the added value of this approach in sheep infected with Bluetongue virus and chickens infected with Highly Pathogenic Avian Influenza virus. In the sheep experiment, the accelerometers detected activity drops one to two days earlier than human observers and confirmed the clinical symptoms in the treatment group (as compared to controls) also from a behavioural perspective. Moreover, we found in both sheep and chickens that lowering the sampling frequency from 25 Hz to 1 Hz had a neglectable effect on general VDDBA trends while massively extending battery lifetime, suggesting that 1 Hz could be sufficient in this context. Currently, we are implementing a real-time tool using Bluetooth Low Energy accelerometers with edge computing. Overall, we recommend wider implementation of sensor technologies in animal trials to improve the quality of experimental results and refine animal experiments.

Self-contained unit for remotely monitoring bison: ID, performance, welfare and health cues

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North America Bison (*Bison bison* subsp. *bison* or subsp. *athabasca*) are wildlife currently utilized as indigenous livestock, commercially raised for meat. While bison can be successfully managed on rangelands and in feedlot settings, this species is adversely impacted by excessive handling. As a result, limited opportunities exist to routinely access animal performance. The aim of this project was to develop a non-invasive self-contained unit to enable remote assessment of bison performance. A walk-through unit enabling single file passage of bison was developed and deployed between feed and water sources to encourage continual throughput of animals. The solar and wind powered unit was equipped with an RFID ear tag reader for animal identification, a walk-over-weight scale, a weather station, and thermal and visual cameras equipped with laser sensors to trigger activation. The system was controlled remotely, enabling multiple sampling protocols and data acquisition and transmission was facilitated via an onboard processor connected by LTE to the cloud. The thermal and visual cameras images of both the face and the entire dorsal body position of each animal were taken. During each passage through the unit, information about animal ID, body weight, images and concurrent weather data was collected. The continuous assessment of body weight enabled detailed assessment of growth curves. Visual images were used to assess the body condition score of each animal based on curvature of the body near the tail-head viewed dorsally. The thermal images further supported the assessment of body condition, as well as provide additional information on animal metabolic rates and febrile early disease detection. Weather data provided environmental information to correlate with and correct animal measurements. The successful deployment of this unit negates the need for handling bison for routine procedures such as weighing, and additional data streams enable continuous monitoring of husbandry practices to ensure improved animal welfare and respective performance outcomes.

Session 56

Theatre 3

Detection of multiple feeding behaviours in calves – a comparison of sensor positions and window sizes

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The use of wearable accelerometer sensors to capture livestock behaviour is promising and keeps evolving. The current investigation sought to evaluate the positioning of triaxial accelerometer sensors and the impact of window size in classifying multiple feeding behaviours in calves. Out of 10 calves in an original experimental setup, three animals, all of which were fitted with triaxial accelerometer sensors at both the area of the right cheek and in the neck were followed for the current analysis. Besides the recording of sensor data which lasted for three days at a frequency of 20 Hz, direct continuous focal observations lasting for about 10 hours per animal were also made and, activities including feeding, rumination and suckling were recorded. Furthermore, the sensor and observational data were merged and five different datasets were generated according to predetermined window sizes: 1 s, 5 s, 10 s, 30 s and 60 s. Each dataset consisted of 19 features extracted by computing simple statistics of the raw accelerometer variables and the magnitude of acceleration. For each animal, separate analyses were performed for each sensor position and window size specification applying both random and bootstrap forest approaches. Additionally, we analysed the scenario that combines features from both sensor positions. The combined-feature analyses produced the highest mean accuracy (90%, 60 s) while there was no discernible difference in mean accuracy between the neck and cheek positions. However, there was evidence of an impact of window size variation. Sensitivity was highest (> 90%) for rumination and suckling, with 10 s window size and sensors at the cheek. Specificity for the three feeding behaviours were generally high (92% – 99%). Features such as the mean and interquartile range of the magnitude of acceleration contributed largely to the accuracy of prediction. For practical and animal welfare reasons, the attachment in the neck is more suitable, as only a collar needs to be worn by the calf instead of a halter. Our results provide a valuable insight into the setting up of an automatic behavioural detection system for calves.

Early prediction of declining health in small ruminants with accelerometers and machine learning

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Assessment of the health status of individual animals is a key step in timely and targeted treatment of infections, which is critical in the fight against anthelmintic and antimicrobial resistance. The FAMACHA scoring system has been used successfully to detect anaemia caused by infection with the parasitic nematode *Haemonchus contortus* in small ruminants and is an effective way to identify individuals in need of treatment. However, assessing FAMACHA is labour-intensive and costly. We used accelerometers to measure the individual activity of extensively grazing small ruminants (sheep and goats) exposed to natural *Haemonchus contortus* worm infection in southern Africa over long time-scales (13+ months). When combined with the Support Vector machine learning algorithm, this activity data can predict poorer health (increases in FAMACHA score), the sheep farm prediction AUC was 84% and 68% for the goat farm. We also demonstrated that using exogeneous data such as rainfall which is known to be directly linked to *Haemonchus contortus* worm infection yields lower or no predictive power than using accelerometer data, 74% and 50% AUC on sheep/goat farm respectively. Interpretation of trained classifiers reveal that poorer health affects the night-time activity levels in the sheep. Our study thus shows that low cost biologgers can detect subtle changes in animal health and enable timely and targeted intervention. This has real potential to improve economic outcomes and animal welfare as well as limit the use of anthelmintic drugs and diminish pressures on anthelmintic resistance. Our work here relies on data collected with a close source sensor that presented multiple challenges including data access and quality. To further close the gap between academic research and real-world applications we decided to develop our own open source programmable bilogger. Our work is available as a preprint at <https://www.biorxiv.org/content/10.1101/2020.08.03.234203>

Session 56

Theatre 5

Prediction of behavior activity of donkeys at pasture using data from a tri-axial accelerometer

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Monitoring behavior of grazing animals can be important to manage animals at pasture. With this aim, data collected with a tri-axial accelerometer device was analyzed, using a multivariate approach, to discriminate the three main activities (grazing, walking, and resting) of donkeys at pasture. A total of eleven donkeys were equipped with the accelerometer tied to the collar. The device recorded, at each second, acceleration values according to the three cartesian directions. The behavior of the involved animals was also video recorded. From the original data, five datasets were obtained by fixing time-epochs of 15, 30, 60, 90, 120, 150, and 180 seconds, respectively. The raw acceleration data was processed for each time-epoch to create 12 variables: the mean, variance, and inverse coefficient of variation for the three axes and the resultant. The canonical discriminant analysis (CDA) was exploited to discriminate between the three behavioral activities and to assign time-periods to the correct behavior. CDA significantly separated the three behavior activities (Hotelling's *t*-test < 0.001) for all time-epochs. The best result in assigning time-periods was obtained for 120s and 150s with a total classification error around 9% for both. In particular, among behaviors, eating and resting showed the lowest (4%) and the greatest (17%) error, respectively. Similar results were obtained for 60s and 90s with a classification error of 10%. The worst classification was obtained using the time-epoch of 15s which showed an error of 13%. Data from these systems can help to understand donkeys' behavior at pasture. This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PNRR INVESTIMENTO 1.4 -D.D. 1032 17/06/2022, CN00000022)

Monitoring of nest use through RFID tracking in cage-free laying hens

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This study evaluated the frequency of nest use and behaviour inside the nests of brown hens housed in an aviary system. A total of 225 hens were individually tracked through a passive radio frequency identification (RFID) system. The accuracy of the RFID system in identifying animals entering/exiting the nests and the behaviours of hens (exploration, preening, rest, and deposition) inside the nests were assessed through video recordings (20 minutes every two hours) performed at 21 and 28 weeks of age, in two separate nests and two days a week (4 days of recording). The number of hens inside the nests, regardless of age, was strongly influenced by the hour of observation ($P < 0.001$), with the highest number of hens recorded between 2 and 4 hours after the opening of the nests. The time spent inside the nest did not change with the age or the hour of observation, whereas the exploration behaviour changed with the hour of observation ($P < 0.001$). The correlation between RFID and video-recorded data ($R_2 = 0.47$) suggests a good estimation of the number of animals inside the nests at a given time based on the RFID system. In conclusion, this preliminary study highlights the potential of the use of RFID system to monitor nest use in laying hens in an aviary system. Acknowledgements: This study was carried out within Agritech National Research Center and was funded by the European Union – NextGenerationEU (Piano Nazionale di Ripresa e Resilienza (PNRR) – Missione 4 Componente 2, Investimento 1.4 – D.D. 1032 17/06/2022, CN00000022).

Session 56

Theatre 7

ModBehav, a supervised classification algorithm to predict goat behaviour from accelerometer data

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Individual animal behaviour monitoring can provide an early sign of variation in animal welfare. Artificial Intelligence and sensors are promising to predict animal behaviour automatically. This paper aims to present a pipeline using a supervised classification algorithm called ModBehav for automatically predict certain animal behaviours from accelerometer data. This pipeline is designed to be generic and applicable to different species, behaviours and types of accelerometers. It was applied on data obtained from 8 indoor-housed goats equipped with ear-mounted accelerometers. “Ruminating”, “head in the feeder”, “lying” and “standing” were continuously sampled from camera recordings for 11 hours for each goat to evaluate the model performances, using the AUC score (Area Under the Curve). AUC score plots the True Positive against the False Positive rate across various threshold values, when common metrics such as accuracy, sensitivity, specificity and F1-score use a fixed-threshold. AUC score provides a reliable metric to identify which model is better on average. For each behaviour, various filtering techniques, time-window segmentations, additional time-series data, and feature selections were tested. The best processing treatments were selected based on the highest AUC score obtained. “Ruminating”, “head in the feeder”, “lying” and “standing” were best predicted with AUC scores of 0.792, 0.806, 0.828 and 0.832, respectively. For all behaviours, a decrease in the AUC score was observed when the prediction was made with data from goats that have not been used during the training of the model.

Exploring the retention of learning in dairy cows exposed to Virtual Fencing after 90 days of non-use
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Virtual Fencing (VF) is an innovative technology for grazing management. In VF system the physical boundaries are replaced by virtual ones, and the containment of animals is ensured by the delivery of 2 deterrent stimuli: an audio tone (AT), followed by an electrical impulse (EI) if the animal continues to move away from the virtual boundary. It has been shown that animals learn the system within 2 to 4 days. However, it is unclear whether they can retain this learning after a period of non-use. A mixed dairy herd consisting of Grigia Alpina (n=14), Pezzata Rossa (n=3), and Jersey (n=4) lactating cows was managed with VF collars on a lowland pasture for 11 days as learning period (L), changing virtual boundaries daily. The animals were then moved to a mountain pasture for 3 months, without using VF. This was followed by a VF re-learning period (RL) of 11 days, following the same scheme of L period. The study aimed to evaluate the retention of the learning capacity of the animals by assessing the differences in the emission of AT and EI between the two periods (L vs. RL). The data were analysed using generalized mixed-effects models. The results showed that the total number of both AT and EI was significantly higher during the L period respect to RL period, with a high probability of receiving an EI during the first 8 days. In conclusion, the results demonstrated that animals could retain learning after a period of non-use, confirming the flexibility in the using of the VF.

Session 56

Theatre 9

Rumen pH changes over a lactation of dairy cows in commercial farm environments

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Current feeding systems for commercial dairy herds aim to increase efficiency of feed used by formulating energy dense diets. However, differences in rations and management, place challenges and can impair the animal's health. It is thought, for example that SARA is common occurrence. Several diagnostic methods have been developed, however these are invasive, identification of affected animals is challenging and information in non-fistulated animals and across the different stages of lactation is scarce. Over the last few years new technology has been developed; amongst which intra-ruminal boluses present a viable alternative. Therefore the aim of the present study was to collate rumen pH data from lactating dairy cows under commercial farm environments. Four trials were carried out at the University of Edinburgh, UK during 2012 to 2015. Trial (T) 1, 14 multiparous (multp) cows days in milk (DIM, mean \pm SEM 104 \pm 12 d) and parity [median lactation (L) = 4], T2, 14 multp cows (DIM 97 \pm 6, L = 4), T3, 14 multp cows (DIM 139 \pm 4.5 d, L = 4) and T4, 34 cows (13 heifers, 7 2nd lactation and 14 3+ lactations). All cows were orally administered a pH bolus (WellCow Ltd.,UK) and managed as standard farm practices: offered TMR and concentrate fed to yield, in T3 cows had access to grazing. All statistical analyses were carried out using R. Feeding and management practices had no effect on performance parameters amongst Trials which allowed the evaluation of rumen pH data. Table 1 shows collated rumen pH data. Rumen pH values recorded across lactation represent high variability in rumen pH values observed, with ranges of more than 1 pH unit across lactation amongst individuals kept under the same conditions and consuming similar diets. Our results show a novel insight of the rumen pH dynamics throughout lactation: the transition and early lactation stages, were the most challenging, possibly due to the physiological and environmental stress faced by the animals in said periods. Further work is warranted to further investigate rumen pH in more diverse and challenging environments

Integrating Infrared Thermography to Assess Thermal Stress and Behavior in Dairy Goats

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Infrared thermography (IRT) offers a non-invasive approach to monitor superficial skin temperature (SST) in livestock, yet its application in goats remains understudied. We investigated 18 Alpine dairy goats over six days (July/August 2023) during the two daily meals (at 7 AM and 7 PM) to determine the association between Temperature Humidity Index (THI) and changes in goats' behaviour and SST across various body regions (eye temperature and SST of the flanks, back, knee, and distal leg). The goats were video recorded for 10 minutes each time and behavioral assessments were conducted using an ad libitum sampling method. Temperature and humidity were recorded using a weather station, and THI was used to categorize heat stress into acceptable ($THI < 75$), moderate ($75 \leq THI \leq 80$), and severe ($THI > 80$) based on previous studies. Distal leg and back SST exhibited the strongest association with THI variation (adjusted $R^2 = 0.88$, $p < 0.001$ for distal leg and back SST). Furthermore, goats experiencing moderate and severe heat stress exhibited reduced rumination while standing ($p < 0.001$), less self-grooming ($p = 0.031$) and higher SST for all body regions ($p < 0.01$). The integration of IRT in thermal stress assessment presents a promising avenue for precision livestock management, enabling early detection of thermal discomfort in goats. Acknowledgements: This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022).

Session 56

Theatre 11

From MIR predicted fatty acids in bulk tank milk to a herd health monitoring tool for farmers

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Previous research has established associations between the fatty acid (FA) profiles and parameters such as energy balance, heat stress and dietary composition. Consequently, FA profiles predicted from mid-infrared (MIR) spectra of bulk tank milk hold promise as valuable indicators of herd health. Leveraging routinely collected data from bulk tank milk, this study employs unsupervised learning techniques on a large dataset ($N = 803,411$) to cluster herds based on their MIR-predicted FA composition, thereby assessing herd status. Seven distinct clusters were identified, with one exhibiting associations with health issues, while at least two others were linked to management practices such as grazing. The health issue cluster has the lowest predicted milk yield, energy balance and protein efficiency, while having the highest predicted blood β -hydroxybutyrate and blood non-esterified fatty acid. These clusters were predicted from FA features using the random forest (RF) algorithm, achieving a cross-validation accuracy of 92% but also directly from spectra with an accuracy of 76%. By analyzing the dynamics of herds over time using these predicted clusters, we observed that herds categorized as having a "healthy" status never transitioned directly to a "sick" status; instead, they moved through an "intermediate" status. These intermediate stages serve as promising indicators to assist farmers in monitoring herd health using routinely acquired MIR data.

Constitution of an international dataset on blood biomarkers in dairy cows: a preliminary study to develop milk MIR models

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Blood composition remains the gold standard to monitor and detect various health disorders of dairy cows. Estimating blood components through non-invasive methods would enable to scale up the measures in terms of cow number and time frequency, while aligning with societal requirements. This work begins with the constitution of a large dataset from multiple organizations across 12 countries. The first objective was to conduct an explanatory study, to better understand the variability of blood biomarkers regarding animal characteristics, sampling protocols and their relationship with other phenotypes of interest. The second objective was to improve on the large variability to develop robust models based on milk MIR spectra. Data merging resulted in a dataset of approximately 10,000 individual records of blood reference values and associated milk spectra. The majority of records were associated with blood BHB and NEFA, and fewer records with glucose, IGF-I, fructosamine, cholesterol, urea, progesterone, calcium and phosphorus. This preliminary work will facilitate a better understanding of the sources of variability in biomarkers, to highlight optimal modelling methodologies among linear and non-linear algorithms, and to estimate the capacities of milk MIR spectra to provide information on those traits under routine conditions

Session 56

Theatre 13

In-line milk analysis: how accurate can we get?

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As milk contains valuable information on the cow's metabolic status, regular analysis of the produced milk is a very efficient way to monitor cow and udder health. Near infrared (NIR) spectroscopy has been shown to be valuable for rapid, non-destructive and on-line analysis of the raw milk composition. In the past 15 years, several prototypes for in-line milk analysis have been developed and validated, some of which are commercially available nowadays. Still, none of these sensors got ICAR certified so far, generally indicating that the accuracy, repeatability and reproducibility are insufficient. Based on our own 15-year experience, we will present and discuss the main challenges for in-line milk analysers by reflecting results and sensor performances reported in the past against those obtained with our own research-prototypes for in-line milk analysis. These prototypes have been extensively tested at different dairy farms and under varying conditions over the past 7 years. Although the initial (calibration) accuracy of the prediction of milk fat, protein and lactose is largely depending on the signal-to-noise ratio of the sensor itself, the predictions afterwards are subject to drift that is mainly characterized by a bias. Part of this bias drift comes from variation in milk temperature, which follows a seasonal pattern and can be accounted for by robust modelling. Additionally, unsupervised techniques and using the bulk milk analysis to correct for this bias can help and bring the performance of the sensor within the required specifications. Finally, it was found that sensor maintenance and good cleaning and milking practices are crucial for optimal sensor performance.

A Deep-Learning Model to Identify Call-type and Individuality of Dairy Cattle

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In this study, we used deep learning (DL) techniques for developing AI models for classification of high- (HF) and low-frequency (LF) calls, and for individual cow identification, based on 1144 vocal recordings of 20 adult cows during visual isolation from herd-mates. The dataset contained both HF (n=952) and LF (n=192) calls, with PraatDSPv.6.0.31 package being used for the extraction of 23 acoustic features/vocalization. The DL framework used a deep-gated recurrent unit (GRU) neural network (NN) model, combining a two-dimensional convolution NN and recurrent NN, based on the GRU cell unit that gets as input the spectrogram of the audio signal. The training-set contained 80% of the vocalizations, while the remaining 20% were included in the testing-set, results were expressed as mean and SD for k=5 fold. The HF and LF vocalizations DL classifier models' performance reached 89.4±3.8% accuracy. The cow's individuality DL identification classifier models' accuracy based on HF+LF reached 72.5±4.7% accuracy. These results demonstrate the effectiveness of deep learning techniques in classifying HF and LF calls, as well as for identifying individual cows based on their vocal productions.

Session 56

Poster 15

TechCare: Monitoring semi-intensive meat sheep welfare through EID and weight

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Managing the welfare of sheep in grazing systems can be challenging but may be improved by the use of precision technologies and digital tools to facilitate monitoring. The TechCare project has investigated the use of commonly available tools on farms (electronic identification (EID) which is mandatory in the EU and EID-linked weight crates) to provide welfare-related information. The aim of this study was to determine if data obtained from EID and weight could act as a warning of potential welfare issues in UK meat sheep, including gastrointestinal parasites, lameness, mastitis and undernutrition. Field studies were conducted at 2 sites in Scotland. At site 1, 30 ewes and their twin lambs grazed fields from April (lambing) until October over 2 years and were naturally exposed to welfare challenges, while at site 2, 100 ewes were monitored over the summer and winter on rough grazing ground. Every 2 weeks, EID, weight, and welfare scores using the AWIN protocol were recorded. At Site 1, worm faecal egg counts were also recorded from each ewe and lamb and behavioural observations were taken five times per week. Results show that the main welfare issues were lameness in both sites and age groups and that mastitis in ewes and gastrointestinal parasites in lambs were common at site 1. Weight recording using EID-linked weigh heads was robust at both sites. Weight was affected by level of gastrointestinal parasites present at site 1. The potential of EID and weight to provide information about welfare will be discussed.

Labour: a key issue for the long-term future of French equine businesses

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The equine labour market is under pressure, with 15-20% of jobs offers being renounced for lack of a suitable candidate. Recent work carried out by the Equine Network has shown the tight situation of these equine businesses, which have little room for manoeuvre in terms of time "I have to manage everything in a hurry", and which also report physical and mental hardship "I'm unsettled... I'm very tired". 74% of the farmers questioned felt they had a heavy workload, and 60% mentioned a lack of free time and a poor work-life balance. "Improving working conditions" was the first need mentioned by French equine farmers as part of the EUnetHorse project on the resilience of equine farms. These initial findings highlight the extent to which the issue of work is vital to the sustainability and appeal of these professions. Solutions need to be found to improve working conditions for farmers and to encourage young people to think about their projects. DECLIC TRAVAIL is a free online platform developed by the French Livestock Institute in 2020. Its role is to raise awareness of work issues among livestock farmers of all types, including equine farmers. It enables them to carry out a self-diagnosis and to think about possible solutions to improve their work. The platform offers over a hundred solutions sheets focusing on three issues: working conditions, human resources and work organisation. The equine sector was integrated into the platform in 2024 and offers around forty work solutions to equine farmers. If they are unable to find satisfactory solutions to their problems, farmers have access to a list of advisers specialising in their sector. To conclude, this tool is designed to raise awareness of the issue of work and offer solutions to improve the operation of the business. Improving working conditions will increase attractiveness of this profession.

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Theatre 2

Self-organized equestrian users: towards services adapted to the demand

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Self-organization is increasing in sport and recreative activities and a mismatch between the service offer and the consumer demand is often observed. Following this trend, more and more French equine owners organize themselves to take care of their equine outside any professional equestrian structure. Despite the health, safety, regulatory, economic, social, and environmental issues related to them, little is known about these consumers. In this context, a research program was designed to analyze self-organized equestrian user characteristics, practices, needs, relations with professionals... More precisely this communication aims to compare the demand of these consumers (and their willingness to pay) with the interest of professionals in offering these services, and at what prices. The methodology relies on two surveys conducted online in 2023, one among 337 self-organized equestrian users, and another among 233 managers of professional equestrian structures. Results highlight a gap between the offer and the demand. Professionals often consider these customers as irregular and occasional, whereas 90% of self-organized respondents have needs for services. This demand is divided into four main categories: daily care and management of equines, equestrian practice, agricultural services, and rental of specific infrastructures. Finally, comparing actual and potential supply with the demand makes it possible to highlight examples of typical services in each of these categories, associated with prices matching supply to demand. Our conclusions highlight the development potential of this market, which still remains to be invested in order to better respond to self-organized equestrian user needs and to better support them.

Riding teachers can affect horse and rider biomechanics in the design of their lesson

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Instructing riders is a complex task, the teacher needs to pay attention to both horse and rider, while the rider needs to listen to both teacher and horse. The aim of this project was to investigate the interaction between teacher, rider and horse in relation to instructions and timing within instructional spaces where the rider asks the horse to perform a transition. The study included five riders and two riding teachers in Sweden, and ditto in Norway. Each rider received instructions on two horses, in total 40 lessons. The riding teachers were interviewed regarding their teaching. All lessons were video recorded and analyzed using conversational analysis. Horse and rider kinematics and rein tension data were collected and analyzed in mixed models. Results shows that the teachers thought it was important to make room for discussion and to give quick and timely feedback to help the rider understand and perceive what to strive for. The teachers designed their lessons differently. Two teachers spent about a third of their lessons preparing the riders, in walk respectively by riding on a circle, while two began with straight-line walk-trot transitions early on. Maximum rein tension before and during down-transitions was lower for the riders that spent more time on preparation. Two teachers focused more on the walk and two focused more on the horse to be quick and attentive. Focusing on the walk, by asking the riders to count each walk stride between down- and up-transitions, resulted in a walk where the relative timing of the up-down movement of the withers and croup was closer to the ideal. When asking for longer walk sequences in-between transitions and commenting on rhythm the riders achieved a walk with better hind limb protraction consistency. The results highlight the importance of preparation and lesson design and show that an experienced riding teacher can have an influence on riders' development of a skill, as well as a measurable effect on rein tension and the quality of the walk. Experiences gained in the study can be used to improve the effectiveness of teaching riding to benefit both riding and horse welfare.

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Theatre 4

Farms with Equines in French Agriculture: Reflections on the Evolution of the Equine Sector in the 21st Century

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After 1945, the presence of equines sharply declined in the agricultural sector due to mechanization. Revival plans during the latter half of the 20th century aimed to preserve the main native horse breeds for meat production and tended to develop breeding for saddle horses and ponies, whereas the racing sector sustained itself through betting. At the beginning of the 21st century, the question arose regarding the role of equine production in agriculture: whether it should primarily serve human consumption or diversify towards leisure activities, potentially including high-value-added productions for equestrian competitions and races. To address this question, we analyzed farms with equines (FE) in the last censuses (2000, 2010, 2020), distinguishing main types of equine herds. While the national equine herd remained relatively stable at around one million heads over the past 20 years, census figures declined: from nearly 450 to 312 thousand heads, and from 81 to 27 thousand FE between 2000 and 2020. However, these figures were also due to stricter criteria of the 2020 census, mainly affecting smaller structures and those with few or no equine births. So smallholders (1 to 3 equines), representing 63% of FE in 2000, accounted for only 40% in 2020. However, the distribution of FE by type of breeds with 4 or more equines remained relatively stable: the saddle and racing sectors remained largely dominant across censuses, comprising over 2/3 of FE, while the draught sector fluctuated around 15%, and donkeys accounted for 4 to 6% of FE. These relative weights reflected national features of the French horse industry (IFCE, 2024). To better understand the 2000-2020 trends, we analyzed FE trajectories (based on SIRET numbers). As 2/3 of 2020 FE were already recorded in 2010 (46% with equines, 20% without equines), 34% were not recorded before and were mostly specialized in saddle and race horses. These findings showed that the equine sector in 2020 was primarily driven by "young" farms or those that acquired horses within the past decade, indicating that this predominantly non-food sector is expanding within the agricultural sector.

Income prediction in riding schools and boarding stables in France – 1st assessment

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Amidst an environment of inflation, we have devised a methodology aimed at estimating income generated by equine farms. This method amalgamates purchase price indices for agricultural inputs (IPAMPA) with field expertise. The income projections hinge on data sourced from a panel of equine farms within the "INOSYS" Equin network, drawing from a consistent sample comprising riding schools and boarding stables. The aim is to estimate the level of income earned by horse farms over the past year. Cost Projections: Annual fluctuations in input prices, goods, and services are gauged in accordance with national trends derived from IPAMPA data (<https://idele.fr/ipampa>). IPAMPA serves as a comprehensive index tracking variations in prices of goods and services pertinent to agricultural activities. Updated on a monthly basis, it sources data from four primary outlets. Expenses specific to equine activities are selected based on these datasets. Sales and Assistance Projections: Sales figures are derived from the accounts of the monitored equine farms. These encompass the selling prices of products and their corresponding volumes, as recorded within the INOSYS system by engineers from the Chambers of Agriculture, the National Federation of Horse Councils (FCC), and the French National Horse and Riding Institute (IFCE). Expert assessments determine the percentage changes in service prices and sales volumes based on field surveys. This work serves as a valuable resource for the equine industry and policymakers alike. The annual estimation of equine farm income is compiled at the outset of the calendar year (February), following the collation of all IPAMPA indices. It furnishes an overview of the financial health of equine farms in the region, presenting current income figures in euros. This work provides a trend in the economic results of the farms. Nevertheless, forecasting income on horse farms is a difficult exercise, given the diversity of activities on each farm and the fact that our samples are very small.

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Theatre 6

Lessons Learned from Horses for School Practice

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Lessons Learned from Horses This study examines whether the well-being of special class students could be influenced by optional interventions. Finnish national curriculum emphasizes students' active involvement, a joy of learning and positive relationships. Thus, all three interventions had co-operational and social skills tasks. Equine-assisted social education (EASE) is an informal intervention with non-verbal equine activities. Aggression Replacement Training® (ART) is a behavioral program for social skills and anger control. Co-operational mathematics (MATH) is an approach to collaborate on mathematical and problem-solving tasks. These interventions are conducted eight times during the spring semester. The results of each intervention group were compared with each other as well as with a treatment-as-usual (TAU) control group. The primary instrument is the Achenbach System of Empirically Based Assessment (ASEBA) collecting data on students' school success and well-being. Parallel instruments measure the aggression, the empathy, the locus of control and the loneliness. 60 students from regional special classes participated in this study, 27 of them were on the third grade and 33 were on the seventh grade. Due to a small sample the analyzes are conducted with non-parametric tests. Boys who participated in EASE improved withdrawn-depressed symptoms, somatic symptoms, social problems, attention problems, and aggressive behavior. However, both EASE-groups boys and MATH-group boys had the lower internal locus of control in the post-test assessment. The ART-group had improvement in thought problems and rule-breaking behavior. Students in control group improved with lower withdrawn-depressed symptoms and thought problems empathy.

Composting as a sustainable way of utilization of horse manure

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An appropriate utilization of horse manure poses major challenges for numerous horse farms. The high proportion of straw in horse manure and its unknown hygienic status reduce the suitability as a fertilizer, especially for use in grassland. Urgently, sustainable ways of using horse manure are needed. The study analyses the eligibility of composting horse manure with subsequent use as fertilizer. Within the scope of six-week trials, four differently treated muck heaps were investigated. The examination focuses mainly on the temperature-related inactivation potential of pathogens, the influence on the degradation of anthelmintic residues as well as a reduction of the manure volume. During the trials, temperatures of over 75 °C were measured in the center of the chopped muck heaps. Temperatures in areas near the ground are significantly lower and clearly oriented towards the ambient temperatures. The general temperature level in the chopped muck heaps was higher than in the control heap. The study indicates that a reduction of salmonella by more than 4-log levels is principally possible, but not at all positions in the muck heaps. The absence of a sanitization was mainly observed in areas of close to the ground. Consequently, muck heaps should be turned to mix the material and ensure a sanitization. By chopping, the straw-based manure prior to composting, up to 65 % of required storage capacity can be saved, compared to the non-chopped manure. A further volume reduction was achieved through the composting process, whereby the reduction potential is higher in chopped manure than in untreated horse manure.

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Theatre 8

Nice to know you – Effects of human expectation on horse behaviour depending on the human-horse familiarity – preliminary findings

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The domestication of horses is more than 6000 years ago – a long time for the evolution of the human-horse bond. Understanding the interaction between humans and horses is an important issue for animal welfare and human safety. In this study we replicate a study design of Keeling et al. (2009) testing the effects of unintentional signals given by the human to the horse with the focus on the behaviour of horses. Further, we analyze the difference between the reactions of horses with regard to the familiarity between horse and human. The humans were asked to lead their horse on a defined track in an indoor arena. The track had to be walked four times. Before the test, humans were told that there would be an umbrella opening in the fourth round, without actually putting this into practice. Each horse (n=7) had to fulfil the task twice, the first time led by the owner, the second time led by an unknown person. The behaviour of humans and horses was recorded and analyzed. Data (types of behaviour) was analyzed using linear mixed models including familiarity (known/unknown), round (1-4) and the interaction between both as fixed factors, the horse was included as random factor. We found no significant differences between rounds. However, the familiarity revealed a significant effect on some of the parameters observed (e.g. length of the rein, head shaking in the horse). No effect of the interaction between familiarity and round could be found. In summary the study could confirm that, even if the expectation of the human revealed no effect, the familiarity between human and horse is reflected in their behavior, which might be relevant for further studies on the horse-human interaction.

EUNetHorse - setting the base: results on characterization questionnaires at EU level

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EUNetHorse is a Thematic Network EU project funded under the Horizon Europe program. This project aims to improve the resilience and the performance of equine farms in Europe by providing a network of exchange of needs and existing solutions and good practices promoting exchanges between actors of all levels (regional, national, European), of all types (professionals, advisors, trainers, researchers, policies, etc.) and of all sectors (sport, leisure, work, milk-meat, races, tourism, etc.). The first step was to characterize the equine sector under the activities of the project. In this way, 40 interviews of farmers were conducted in the 9 countries of the project (Belgium, Finland, France, Portugal, Romania, Spain, Switzerland, Germany & Poland) to characterize at country level and later at EU level the population involved in this project, and to identify needs and best practices. For the purposes of the project an equine farmer is a professional who breeds or keeps horses while at the same time adds value to the utilized agricultural area. This condition had to be fulfilled at least in 80% of surveyed farmers. The aim was also to have the widest possible representation of the equine sector in each country. A total of 462 interview interviews were performed in all the involved countries. Average age of farmers is 48 years, and 57% of farmers work full time in the area. The EU average number of horses/farmer is 45 with a median of 25 animals (maximum 525, minimum 1), with an average of 10 foals/year. The average area/farm is of 138 hectares, with a total of 41 094 hectares in the interviewed farmers, of these 31% are dedicated to permanent meadows for the horses, and the rest to areas for forage production. Concerning social aspects, the average number of workers in farms is 5 and the distribution is 57% male and 43% female. These preliminary results allow to draw a first generalized characterization of EUNetHorse universe.

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Theatre 10

Italian heavy draft horse management: practices and welfare issues

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The Italian heavy draft horse (CAITPR) is becoming a growing reality[1]. Given the different roles this breed plays, including meat production and sport, it is pivotal to understand its management practices. This study aims to investigate the practices and welfare issues related to CAITPR breeding in Italy. CAITPR breeders were subjected to a survey from March to November 2023, questioning respondents' details and management practices. The survey generated 92 responses (representative sample of the population) and was completed by breeders from 16 regions, demonstrating the wide Italian distribution of the breed. The respondents were mainly male (85.7%), aged 51 years on average, who reported having 20 years of breeding experience. The majority of them kept horses for more than one purpose (e.g. shows, breeding, work and fattening), highlighting the multiple CAITPR roles. Most horses lived in groups (79.3%), on pastures (41.3%) or in paddocks (34.8%), but 17.4% lived in single stalls. Horses were often fed with hay ad libitum and grains, but 20% lived based only on pasture, thanks to the breed's adaptability to low-nutrient conditions. Concerning health, only 3.3% of horses were not vaccinated, while deworming was always performed. Dental trimming was not a routine practice, while hoof care was often done as needed (31.5%) or once a year (33.7%). The most commonly reported diseases were diarrhea (22.8%), colic (8.7%), and wounds (7.6%). Hot branding was a widespread practice (78.3%), often requested by breeders to identify purebred subjects. Finally, more than two-thirds of horses were trained to the use of the halter. In conclusion, these preliminary results outlined the general practices, highlighting the points that need to be improved to ensure a higher level of welfare for this breed. [1] <http://www.anacaitpr.it/>

To eat or not to eat horsemeat? - a review of cultural, ethical, and practical considerations

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The aim of the study is to analyze the debate on the consumption of horse meat from a cultural, ethical and practical perspective. Experimental methods included a literature review. In some cultures, horsemeat has been consumed for centuries and is considered a delicacy, while in other cultures it is strongly rejected due to animal welfare concerns, cultural taboos or personal beliefs. In addition, controversies surrounding the mislabeling of horsemeat in food have further complicated the debate. Proponents of eating horsemeat argue that it can be a sustainable and nutritious source of protein, micro and macro elements and vitamins that benefits health and contributes to food security. Indirectly, it increases the profitability of breeding, especially of native horse breeds, thus contributing to the preservation of genetic diversity. They emphasize the importance of respecting cultural diversity and individual eating habits. On the other hand, opponents often raise ethical concerns about the treatment of horses during transportation and in slaughterhouses, as well as food safety issues due to the possibility of mislabeling and the use of veterinary drugs not intended for human consumption. Ultimately, whether or not you eat horsemeat is a personal choice. It is important that each individual considers their own beliefs, values and concerns and educates themselves about the origin, safety and consequences of eating or not eating horsemeat.

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Theatre 12

Production and consumption of horse meat in France: overview and perspectives

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This communication aims to present an overview of the horse meat market in France. It relies on a literature review and a research program whose goal was to understand consumption motivations and barriers, through qualitative and quantitative surveys among horse meat (non-)consumers and chefs. In France, draft foals bred for this market (producing light red meat) are 80% exported, whereas France imports reformed horses (dark red meat) to satisfy 80% of the national consumption. French horse meat consumption is currently declining significantly. Nevertheless, it seems to exist a substantial potential to develop this market for two reasons. First, this meat has nutritional, organoleptic and environmental qualities that are interesting in the current context of a sought for diversification of protein sources. Second, our work showed that out of all French people who do not eat horse meat, 15% would be ready to do so if they had the opportunity, mainly with friends and relatives, or in restaurants. According to the chefs, horse meat is rarely on the menu but is an interesting product for certain types of restaurants. Targeted managerial strategies can be proposed in light of these results, differentiating customers according to their profiles. People with strong moral opposition or having a distant relationship with meat in general mustn't be targeted. However, people who already consume it or those who are open to it would be sensitive to a better availability and visibility of the offer, for consumption at home, but also in commercial catering that appears to be relevant to familiarize new potential consumers with this product. Moreover, it appears important to enhance the qualities of this product, which can meet the current challenges of food sustainability.

Study on winter indoor jumping championships

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The aim of the study was to analyse the indoor showjumping competitions over three seasons after the COVID19 collapse in the horse industry in Romania. The NF Winter Tour Championship of 2021 (5 events), 2022 (7 events) and 2023 (7 events) was taken in study. Every event was extended to 3 days with a constant number of 18 tests (height from 60 to 140 cm). Effects of year, event and test were analysed. Year had a significant effect ($p < 0.05$) on number of starts, couples with one fault and those with more faults. Thus, the number of finished rounds were 1719 in 2021, 3502 in 2022 and 3103 in 2023. Out of these, 42 to 48% were finished without any penalty ($p > 0.05$). Couples finishing the tests with penalties increased from year to year. Event had a significant effect on all observed results ($p < 0.05$). There was a trend that less couples were present at start (from 37 to 25) and the faultless evolutions reduced (from 51% to 40%) as the event number and height increased. Considering the rise of couples in the performance hierarchy, we grouped the heights on three levels: beginners (60 to 85 cm height), intermediary (from 90 to 115 cm height) and high level (from 120 to 140 cm height). A significant effect of height was observed in this aspect ($p < 0.001$). Clear rounds varied from 48-63%, to 40-46% and to 29-39% for the first, second and third height group, respectively. Four points penalty was observed in 6-19% in the lowest heights, and in the second and third groups it varied from 17% to 26%. More than one fault per round, comes out as follows: in beginner's 22% to 46%, in the middle heights 26% to 39% and in high levels from 38% to 56%. In conclusion, all studied factors (year, event and test) had an influence on performance results in the winter indoor showjumping competition.

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Poster 14

Portuguese equine production characterization: results from the EUNetHorse thematic network

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Portugal is one of the nine countries involved in the EUNetHorse Thematic Network. This project has as main objectives to improve the resilience and the performance of equine farms in Europe by providing a network of exchange of needs and existing solutions and good practices promoting exchanges between actors of all levels (regional, national, European), of all types (professionals, advisors, trainers, researchers, policies, etc.) and of all sectors (sport, leisure, work, milk-meat, races, tourism, etc.). In order to characterize the equine sector in Portugal and to collect a list of needs, existing solutions and best practices, 40 equine farmers were interviewed. To ensure a wide representation of the sector, 12 of the 18 Portuguese districts were represented in the interviewees, as well as the main breeds that are bred in Portugal (Lusitano, Português de Desporto, Garrano and Sorraia). Interviews focused on professionals who breed or keep horses while at the same time adds value to the utilized agricultural area that include in their work equine production. Preliminary results show that the average farmer has 50 years old and for 27% of them this sector is their full-time business and 60% is their primary activity. The median number of horses per farmer is 52, with 10 foals being born / year, and an average of 248 hectares of farming area (In a represented total of 10.183 ha). In average each farm has 5 workers with 80,5% of male workers, and therefore 19,5% of woman. Portugal is part of EUNetHorse as a country with strong cultural connections to the equine sector. Results allow a general characterisation of the Portuguese equine sector, that will be used to set a group of National needs on R&D and also a comparison with other EU countries from the project. EUNetHorse aims to improve resilience and performance of equine farms, promoting an European exchange between all agents.

The role of locally adapted breeds in the sustainable transformation of the livestock sector: examples from Italy

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Locally Adapted Breeds are defined as breeds which have been in the country for a sufficient time to be genetically adapted to one or more of traditional production systems or environments in the country. According to the Animal Genetic Resources strategy for Europe (ERFP, 2021), a variety of local breeds have characteristics that make them potentially valuable in the provision of a range of ecosystem and cultural services. Considering the increasing threats from climate change, biodiversity loss and other environmental challenges, the characteristics and potential of local breeds should be valued accordingly. On a total of 6370 breeds included in EFABIS database, 2098 are classified as “native” and 622 as “locally adapted”. This contribution aims to discuss the potential role of locally adapted breeds for the sustainable transition of livestock production systems. Some case studies, based on Italian local breeds, will be presented to discuss opportunities and critical points to be considered for the implementation of sustainable breeding programs. The results evidence the need to increase the number of studied traits to strengthen the characterization of livestock breeds, particularly local breeds, in their production environment.

The role of the private (breeding) sector in sustainable transformation: an example of small scale poultry production in Africa

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Poultry constitutes an important economic activity for the rural poor in many African countries. Poultry are generally raised by small-scale farmers in low-input systems to provide nutrition and income to rural households. Often local breeds are used which lack productivity compared to other improved low-input (dual-purpose) breeds such as SASSO. Access to quality genetics can transform the poultry value chain as demonstrated by the “African Poultry Multiplication Initiative” (APMI) in several countries including Ethiopia and Tanzania. APMI has been shown to be an effective model to secure and de-risk the supply chain of quality genetics from hatcheries that produce day old chicks (DOC) via mother units that raise DOC to teen chicks to sell to small-scale farmers. The success of this supply chain is dependent on access to quality dual-purpose parent stock (PS) and operational efficiencies which impacts affordability of teen chicks for small-scale farmers. The private breeding sector can supply 100% exotic dual-purpose PS that fit to these markets. Alternatively, a local male line and an exotic female line can be used as PS, which offers the ability to combine local preferences and adaptability with high reproductive rates to safeguard affordable and improved poultry (example in Burkina Faso). Further, it is key that the breeding program is aligned with the environment of the small-scale farmers. The latter can be achieved by including performance of crossed pedigree birds under challenging conditions in the breeding program (recurrent testing). An effective breeding program, scale, quality and long-term commitment are key for delivering quality poultry genetics. Private industry have the knowledge, global reach and operational efficiency to make this happen.

Anticipating future genetic needs for French dairy and suckler cattle sectors

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The French dairy and suckler cattle sectors are expected to face major challenges over the next 20 years, such as a probable decline in beef consumption, an increase of heat waves, the emergence of new pathogens, constraints on environmental impact, the sensitivity of forage crops to drought, a shrinking number of farmers or volatile input costs. These changes are likely to lead to significant transformation of livestock production systems. Thus, current bovine genetic resources may not be suitable for these future systems and therefore need to be re-investigated. Altering genetic resources being a long-term process, it is essential to think now about the objectives of cattle breeding programmes in order to meet future genetic resources needs. To this end, an interdisciplinary group of cattle experts from various fields (macroeconomics, production systems, ecology, feed production, nutrition, physiology, veterinary sciences, genetics, human and social sciences, public policies) has been set up to define scenarios for the evolution of cattle farming systems in France 20 years ahead, as well as the genetic resources fitting these changes. The expert group identified factors with the greatest potential impact on future cattle farming systems, constructed several plausible cattle farming system evolution scenarios based on the trends of these factors and determined the suitable genetic resources fitting them. Results obtained were then presented and discussed with shareholders of the bovine sector to obtain feedback on the relevance, acceptability and feasibility of these findings. This work was supported by a French government grant managed by the Agence Nationale de Recherche under the France2030 program "ANR-22-PEAE-0003".

Investigating genomic adaptation to climate in local Mediterranean sheep breeds

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Since domestication, livestock followed human migrations, generating several locally adapted breeds. Hence, autochthonous populations often present adaptive traits unobserved in cosmopolitan highly-productive breeds. Using a landscape genomics approach, we investigated genetic signatures of environmental adaptation in 1291 individuals belonging to 63 Mediterranean sheep breeds, genotyped using the Illumina OvineSNP50 beadchip. We used Sam \square ada v0.8.3 to identify genotype/environment associations with 13 environmental variables and correcting for the ancestry fractions identified through ADMIXTURE. Finally, we recorded the genes intercepted by neighboring regions in linkage disequilibrium with each significantly associated SNP. We identified candidate genes of which many have known roles in environmental adaptation. Noticeably, by applying candidate gene prioritization we identified genes putatively involved in metabolic pathways that are particularly relevant, as response to the ever-changing climate conditions (e.g. increasing temperatures and UV-radiations).

Breeding for sustainability: development of an index to reduce GHG in dairy cattle

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Several genetic selection strategies can be incorporated into dairy cattle breeding programs to target a reduction in GHG emissions and provide a mitigation strategy with only modest additional cost, or labour expense to the dairy producer. This can be achieved by targeting genetic progress in a specific trait (i.e. methane) or by building selection indexes that balance economic gain and environmental impact for more conventional traits, or both. Various countries have initiated efforts to incorporate emissions-related traits into their national selection indexes. The strategies for reducing emissions vary due to system-specific objectives and limitations, ranging from specific methane breeding values to broader sustainability indexes. While methane breeding values may not be commercially available in most cases, Canada has taken the lead as the first country to release a methane breeding value and develop a GHG index which includes a direct methane trait. The GHG index proposed for commercialization is expected to reduce emissions per cow per year by 168 kg CO₂e per standard deviation of index and is composed of Herd Life, Feed Efficiency, Methane Efficiency, and Body Maintenance Requirement. The reduction in emissions is largely driven by genetic gain in Methane Efficiency and Body Maintenance Requirements, with results indicating that omission of a direct methane trait from the index would lead to an unfavourable response in individual cow enteric methane output. Motivation for use of a GHG indexes is strengthening in high income countries. This motivation could be greatly accelerated if auditable, transparent and scientifically robust ways of recognising emissions changes due to genetic selection were developed. Ideally, these methods would support both national policy setting and supply agreements with milk processors.

Session 58

Theatre 6

How much animal genetic improvement can contribute to the economic and environmental multiperformance in dairy sheep farming systems?

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Multiperformance is the requirement for a farming system to be efficient from an economic, environmental, technical, and social standpoint. Ruminants largely contribute to greenhouse gas (GHG) emissions and thus there is a strong societal demand for sustainable livestock systems that mitigate methane (CH₄) emissions. It is possible to breed animals producing less methane but effects on other traits of economic interest and on multiperformance at the farm level need to be assessed. This study's objective is to model the contribution of animal genetic improvement to the economic and environmental multi-performance of dairy sheep farming systems. A common dairy sheep farming system in Southern France is simulated using an individual-based model. This mechanistic model of individual sheep is based on the principle of energy allocation between physiological functions to simulate CH₄ emissions and other animal responses to feed variation. The model is deployed at the farm level to simulate individual phenotypic and genetic variability within the flock, according to feeding and reproductive management. Combining mechanistic and genetic approaches allow to examine the influence of both genetic and farm management for their impact on total GHG emissions and the gross margin of the farm. Preliminary results including responses on dairy yield and CH₄ emission are promising but must be validated with real data. These results also need to be extended to total GHG emissions.

Shaping the Future Towards Responsible and Balanced Breeding: Code EFABAR

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The Code EFABAR, the code of good practices for farm animal breeding, delves into the transformation of breeding practices over the past two decades, underscoring the shift from focusing on productivity to including new objectives and traits for a balanced approach. This transition is made possible by the progress in genomics and phenotyping tools and further knowledge of animal genetics, which help mitigate the negative genetic correlations between functional and productive traits. Code EFABAR encapsulates six fundamental pillars of responsible and balanced breeding practices: animal welfare and health, mitigation and adaptation to environmental impact, efficient utilisation of resources, genetic diversity, product quality and quantity, and public and food safety. Adherence to Code EFABAR signifies breeders' commitment to transparency and sustainable practices, prioritising the long-term interests of animals, the environment, and people, including farmers. Code EFABAR offers a comprehensive framework for breeders across various species and farming systems. The code emphasises the responsible use of technologies, focusing on the integrity and well-being of breeding animals when developing and using technologies. It also underscores the importance of management practices and the significance of welfare during transport. To conclude, Code EFABAR is a clear guide for breeders, outlining the principles and practices essential for responsible and balanced breeding. It encourages breeders to engage with stakeholders and the public to promote understanding and acceptance of their work. It serves as a benchmark for the sector, ensuring that breeders meet the highest standards of animal welfare, environmental sustainability, and socio-economic viability. By coactively embracing this standard, breeders demonstrate their continuous efforts towards sustainability.

Session 58

Theatre 8

Breeders' vision to meet future livestock sustainability goals through novel technologies – a Delphi study

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Smart animal breeding can have a central role in the delivery of sustainable livestock production and food security. Emerging transformative breeding and reproductive technologies can help achieve future sustainability goals, but their implementation requires careful consideration of environmental trade-offs and broader societal and regulatory constraints. We used a modified Delphi study to gather breeder opinions on promising technologies for meeting 4 key sustainability goals: high productivity and product quality, high animal health and welfare, low carbon footprint and maintenance of genetic diversity. Panelists consisted of representatives from major breeding companies, breed societies, levy boards and expert advisors of major farmed animal species in the UK. Responses were anonymous except to the lead authors. In round 1 panelists were asked to rank technologies (agri-tech phenotyping, genomic resources, bio-engineering, reproductive technologies, computational tools and prediction models) based on their ability to meet each of the 4 sustainability goals. In the subsequent rounds, panelists were asked to re-evaluate the rank of the technologies considering economic (round 2), and regulatory and societal aspects (round 3). Ranking was based on median (IQR). Summary reports were provided to each panelist after each round. Early results suggest that the ranking depends on the sustainability goal and species. The study provides an insight for scientists, funders, and livestock producers regarding the future use of technologies for next generation farm animal breeding.

Native breeds for agroecological dairy farming: a farmer's perspective

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A transition towards locally adapted and agroecological cattle farming systems asks for a search to suitable animal genetics to support such systems. This may require breeding goals that differ from conventional systems, which could provide opportunities for local breeds. Therefore, we studied the relations between agroecological farming, breeding goals and farmers' perspectives on the value of native Dutch dual-purpose cattle breeds. We selected 34 farmers who mostly keep these breeds in extensive systems. Data was collected for on animal performance, feed composition, land use and farm economics. We visited 15 of these farmers for an in-depth interview about their farming and breeding strategies. The farmers were characterized by their innovativeness and how they experimented with breeding, agroecological practices and business models. They differed in their definitions, motivations for and approaches to agroecological farming and in their associated business models. Farmers commonly indicated to breed for robust, sober animals that thrive on roughage feed. Udder and claw health were also important traits, as well as increasing fat and protein percentages rather than kg of milk. Strategies of farmers to reach their breeding goals differed widely, from using commercial AI bulls to using own bulls. Choices for (crosses with) native Dutch breeds were often motivated by animal health, roughage utilization, beef production and cultural heritage. We will evaluate the interview outcomes with the data of the 34 participating farms.

Session 58

Poster 10

Egg production of local Spanish breeds of laying hens and their physiological stress response to heat stress

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Global warming may adversely affect poultry production. Autochthonous breeds are particularly valuable in a climate change context because of adaptation to the local environment. Differences in daily and cumulative egg production between ten local breeds of laying hens was evaluated in 60 hens per breed for 67 days. In addition, the stress response quantified by the heterophil to lymphocyte ratio (H/L) was evaluated during natural heat stress (23, 26, 28, 30, 34, 38, 40, and 42 °C) in four hens per breed for eight days over a period of 7 weeks. During heat stress, H/L was particularly elevated at temperatures higher than 40 °C ($P < 0.05$). Andaluza Azul and Andaluza Perdiz had high egg production (38 and 35 eggs per hen, respectively) but showed low resilience to heat stress based on their H/L response (0.56 and 0.45, respectively), whereas Villafranguina Roja and Prat Leonada had low egg production (24 and 25 eggs per hen, respectively) but showed high resilience to heat stress (0.31 and 0.30, respectively) ($P < 0.05$). In contrast, Andaluza Negra Barrada had high egg production (36 eggs per hen) and high resilience to heat stress (0.31). Low-input, small-farmer operations may particularly benefit from using local genetic resources of poultry highly adapted to environmental stressors. In addition, crossbreeding of commercial with local poultry breeds may combine the beneficial alleles of high production with improved resilience. Sustainability of production systems in the context of climate change requires novel breed choice and genetic selection objectives for improved heat tolerance.

Combining GWAS via meta-analysis identifies metabolic factors involved in phenotypic plasticity for reproductive efficiency traits

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Reproductive performance is important to guarantee the profitability of the beef cattle production system and genotype-environment interaction (GxE) can affect heifers' reproductive efficiency. Thus, we aimed to identify genomic regions that affect animal adaptation and evaluate the biological mechanisms of candidate genes involved in Nellore heifers' early pregnancy (HP) and rebreeding (HR). For GxE interaction, a two-step genomic reaction norm model via a single-step approach (ssGRN) was used. In the first step, the environmental gradient (EG) was estimated based on BLUE solutions of the contemporary group for average daily gain. In the second step, an ssGRN model followed by GWAS analysis was used for the intercept and slope effect. GWAS results were combined via a multi-trait meta-analysis statistical test. Significant G×E was detected for HP and HR with genetic correlations < 0.80 and animal re-ranking across environments, being more significant for HR. The significant SNP markers Nellore HP and HR were environmentally dependent with a re-ranking of their effects across EG levels. The shared regions between HP and HR for the RN intercept and slope harbored candidate genes related to the gatekeeper for early puberty onset, neuropeptides, growth aspects, and energy pathways, directly affecting sexual precocity and re-conception rate. The GWAS results highlighted differences in physiological processes linked with complex biological interactions between the gonadotropic axes, growth aspects, and sexual hormones to respond to harsh conditions, providing helpful information for selecting adaptable animals with high reproductive efficiency. Acknowledgment: FAPESP 2022/11852-7 and 2017/10630-2

Genotype by environment interaction for milk production traits in Dutch dairy cows in conventional and organic systems

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Genotype by environment interaction (GxE) across animal production systems may lead to re-ranking of the animals between the environments. The EU's Green Deal aims for 25% of agricultural land under organic farming and a 50% reduction in nutrient losses by 2030. This study aims to analyse GxE between first parity Holstein Friesian cows on conventional and organic dairy farms in the Netherlands. The dataset contains 96,527 milk production records from 852 conventional farms and 15,071 milk production records from 224 organic farms. Genetic correlations between conventional and organic farms, calculated for five milk production traits (milk, fat and protein yield and fat and protein content), ranged from 0.95 to 0.99. Hence, there is no evidence for GxE between the two systems. However, there are large differences in farm management within organic and conventional dairy farms. Selecting 50% of the conventional farms with the highest and 50% of the organic farms with the lowest average milk production reduced genetic correlations between both environments to values ranging from 0.84 to 0.89 for the 5 traits. Detailed information from the Annual Nutrient Cycle Assessment is available on these farms and will be used to further characterise farm management. At the farm level, information is available on factors including concentrate use, type of roughage, proportion of cows grazing, and nitrogen and phosphorus efficiency between 2016 and 2021. This will be used to present a more in-depth analysis of GxE between conventional and organic dairy farming systems during the EAAP meeting.

Mining big data to fit animals to climate change, improve welfare and mitigate the environmental impact of livestock productions (BIG Fit Project)

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The generation of big data in agriculture represents a key element toward full sustainability of food production systems. Exploitation of big data in the livestock industry will allow the large-scale investigation of animal genetic make-up, welfare, sustainability of production systems, adaptation to climate changes and farm system. Main issues in handling big data are editing, storage and interpretation. The scope of this note is to present BIG Fit project (funded by Italian Ministry of University-MUR under PRIN-2022 program) that will exploit – for the first time – the largest livestock open linked dataset (2.5 billion records). Big Fit, in partnership with national breeder associations and LEO project is expected to create pioneering breakthroughs in the livestock system. The core project is organized into 2 side WP (project management and dissemination) and 3 core WP (2-3-4). WP2 is carrying out an inventory of existing main data sets harmonizing and merging different data types. Tools for analyzing the data generated by WP2 will be provided by WP3, in which models and algorithms will be developed and tested. In WP4 these methodologies will be then applied to investigate research issues. In conclusion, the Big Fit project has just taken the first steps in order to produce a data inventory. Different data sources are already available: laboratory data and MIR spectra (500 million (M) laboratory parameters recorded on 1.5 M animals), field data (183 M), genomics indexes (131 M), climate and environmental data (11 M) and PLF data (154 M).

Session 58

Poster 14

Genetic parameters of Methane emission in Danish Jersey dairy cows

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The aim of this study was to explore the genetic parameters related to methane emission traits in Danish Jersey dairy cows. Genetic selection for lower methane emission could serve as one of the strategies for mitigating methane emissions in the worldwide transition to greener practices. In this study, we used the sniffer method to measure methane and CO₂ concentrations in the exhaled air during milking of approximately 3000 Danish Jersey cows in commercial dairy herds with AMS milking. Previous research on methane emission in the dairy sector have been explored on Holstein breed only. Therefore, there is a need to study other dairy breeds to facilitate comparisons of breeds and methane reduction potential. In this study, variance components on the genetic parameters of methane- and residual methane traits were estimated using the AI-REML algorithm with the DMU software. Maintenance, production, and efficiency traits such as DMI, BW and ECM, and residual feed intake traits (RFI) were also included in this study. Genetic and phenotypic correlations between all traits were estimated. Results from this study reveal moderate heritability of methane traits as well as moderate to strong heritability for maintenance-, production- and efficiency traits, consistent with previous reports on Holstein cattle. Moderate to high positive genetic correlations were estimated between methane traits and maintenance, production, and efficiency traits; this suggests an incentive for lowering methane production from the global dairy sector by including methane traits in breeding schemes when selecting in the Jersey breed.

Reducing methane emissions through crossbreeding Holstein-Friesian dairy cows with Jerseys

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Reducing enteric methane emissions from dairy systems is important in meeting our climate targets. Crossbreeding may be a viable strategy in achieving reductions. It is well established that crossbreeding with Jersey genetics can improve the efficiency of milk production in pasture-based dairy systems. However, there is currently little information on how this might affect enteric methane emissions. An experiment was established to compare two breed groups, Holstein-Friesian (HF; n = 31) and Holstein-Friesian X Jersey (JX; n = 37). The cows grazed together, managed in a spring-calving grazing system. Methane emissions, milk production and body weight data were collected over 11 consecutive fortnightly periods, running from the 15th of May to the 15th of October 2023. The JX cows had increased daily milk solids production (P < 0.05) despite having a reduced bodyweight (P < 0.001). They also exhibited reduced methane emissions (P < 0.05) and reduced methane intensity (g CH₄/kg milk solids; P < 0.001). Although when expressed per kg of bodyweight, there was no difference in methane emissions between breed groups.

Session 59

Theatre 1

From Waste to Worth: Repurposing Dairy by-products as Source of Innovative Nanosystems

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This study aims to use cow milk whey, the main dairy by-product, as a source of extracellular vesicles (EVs) for biomedical applications. EVs were isolated from whey using a scalable procedure involving centrifugation and size exclusion chromatography. Characterization using nanoparticle tracking analysis, transmission electron microscopy, and Western Blot was conducted. In vitro selectivity assays using MCF-7 (CD44-) and MDA-MB-231 (CD44+) cancer cell lines were performed through flow cytometry (FC) and confocal microscopy (CLSM). Statistical analysis was performed through one-way or two-way ANOVA, followed by Tukey's multiple comparisons (significance set to p-values < 0.05). EVs functionalized with hyaluronic acid (EVs@HA) enabled active targeting of the CD44 receptor on cancer cells. CLSM qualitative analysis showed a higher uptake of EVs@HA by MDA-MB-231 cells, which was corroborated by FC (a 3-fold increase in EVs@HA uptake). Internalization pathways of EVs@HA were shown to be dependent on the CD44 receptor. Overall, this research shows the potential of repurposing whey as a source of innovative delivery systems for cancer therapy, aligning with the principles of circular economy and sustainability. Acknowledgements: FAS thanks her funding from FCT/MCTES through grant [UI/BD/151407/2021], and financial support from BiotechHealth Doctoral Programme. Funding: The authors thank FCT/MCTES for the financial support within the "NanoProMilk" project [PTDC/BAA-AGR/4923/2021].

Nano can be Big(ger): bringing added value to milk fat

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The objective of this work was to produce nanocarriers (NC) from milk-derived components to encapsulate bioactive agents and be used in nutraceuticals. The NC were produced using organic solvent-free methods and vitamin D – which is becoming more deficient with current diets and daily routines – was encapsulated as a model molecule. Developed NC were assessed for dimensions and zeta potential by dynamic light scattering and vitamin encapsulation by UV/vis photospectroscopy. Gastrointestinal resistance was evaluated using biomimetic media and in vitro cytotoxicity using the resazurin viability assay in fibroblasts. Statistical differences were calculated ($p < 0.05$) based on ANOVA followed by Tukey's multiple comparisons test, when data followed a normal distribution (parametric tests); and based in Kruskal-Wallis analysis, when the data did not follow a normal distribution (non-parametric tests). These novel NC presented diameters between 200 and 600 nm, optimal dimensions for oral administration, and zeta potential of -30 mV, maintaining these properties for at least 10 months of storage at room temperature and were loaded with 10% of vitamin. They also resisted both gastric and intestinal digestion, showing no cytotoxicity up to 2.5 mg/mL. Concluding, the developed milk fat-derived NC are exceptional candidates for bioactive delivery applications in the food and medical sectors. The used building blocks bring an added value to dairy fat-based products and promote a circular economy approach. Funding: This work received financial support from national funds (FCT/ MCTES) through the project PTDC/BAA-AGR/4923/2021 'NanoProMilk'.

Standardization method, testing scenario, and accuracy of the infrared prediction model affect the standardization accuracy of milk mid-infrared spectra

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The widespread use of milk mid-infrared (MIR) spectroscopy for phenotype prediction has urged the application of prediction models across regions and countries. Spectra standardization is the most effective way to reduce the variability in the spectral signal provided by different instruments and labs. This study aimed to develop different standardization models for MIR spectra collected by multiple instruments, across two provinces of China, and investigate whether the standardization method (piecewise direct standardization and direct standardization), testing scenario (standardization of spectra collected on the same day or after 7 months), infrared prediction model accuracy (high or low), and instrument (six instruments from two brands) affect the performance of the standardization model. The results showed that the determination coefficient (R^2) between absorbance values at each wavenumber provided by the primary and the secondary instruments increased from less than 0.90 to nearly 1.00 after standardization. Standardization accuracy was significantly affected by the standardization method, testing scenario, and accuracy of the infrared prediction model to be transferred across instruments ($P < 0.05$). This study lays the foundations for developing a national MIR spectra database to be used in dairy farm management and breeding programs.

Assessment of the proteins from whey and its microparticulate and fermented products

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Microparticulation (MP) is a thermal-mechanical process able to denature and concentrate whey proteins, which aggregate in particles with sizes like those of milk fat globules. Microparticulated whey (MPW) is used to produce low-fat products due to its fat mimicking functionality. Recycling whey as a fermented MPW (FMPW) is also a strategy to enhance the sustainability and circular economy of dairy chains. The study investigated the microstructure of WHEY, MPW and FMPW. The whey was bacto-fugated, heated (70 °C, 15 s), ultra-filtrated, microparticulated and then subjected to fermentation with *L. lactis* & *S. thermophilus* vs. *Bifidobacterium lactis*. The microstructure of whey products was analysed in negative staining electron microscopy. SDS-PAGE under both reducing and non-reducing conditions, was also performed with a 4-12% separating gel. Fermentation process showed a limited difference between the two strains since first one highlighted more rapid acidification activity. SDS-PAGE analysis evidenced similar pattern between samples, even if MPW and FMPW had an intense band over 200 kDa that are not visible under non-reducing condition. The difference could be attributed to the modification of inter- and intramolecular sulphhydryl bonds. The native WP displayed irregular or spherical shape with slight aggregation. With MP treatment, number and particle size increased due to aggregation, and numerous small vesicular structures were observed too. This finding can be attributed to the protein interactions at higher temperatures, resulting in the agglomeration of protein particles reaching sizes 1-10 µm, like those of fat globules.

Session 59

Theatre 5

Investigating the effect of total and differential somatic cell counts on bovine milk mineral content

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Improving the understanding of milk quality associated to animal health is crucial for dairy industry efficiency and product quality assessment. This study aimed to investigate the impact of somatic cell score (SCS), differential somatic cell count (DSCC) and their interaction on the variability of seven macro-minerals (Ca, Mg, K, P, S, Cl, Na in micrograms) in bovine milk. Individual milk samples (n = 1,080) were collected during the evening milking in 54 herds of the north-central area of Italy (20 cows/herd). Minerals and somatic cell traits were analysed with an x-ray fluorescence method and a cytofluorimetry-based system, respectively. Content of each macro-mineral was analysed through a linear model that included quintile classes of SCS and DSCC and their interaction, parity and days in milk as fixed factors, and herd as a random effect. Both SCS and DSCC had a significant effect on all the seven minerals. Specifically, milk P, Cl and Na were higher in samples with higher SCS and lower DSCC. The interaction SCS × DSCC affected Na quantity in milk, with higher values associated to an increase of SCS, particularly in samples with low DSCC. Results highlighted the multifactorial nature of milk minerals and the importance of considering different levels of both SCS and DSCC. This study could help to better understand the complex relationships between udder health status of dairy cows and milk quality, particularly in terms of technological and nutraceutical properties.

Multiplex pathogenic bacteria detection in milk with the nanoparticle-assisted porous silicon-based SERS microarray biosensor

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The ongoing threat of milk-related pathogens poses a substantial risk to human health, necessitating the development of efficient detection methods. This study explores the creation of a surface-enhanced Raman scattering (SERS) multiplex biosensor to meet the demand for simultaneous detection of multiple pathogens, thereby reducing measurement time and enhancing food safety. The biosensor utilizes an indirect immunoassay, leveraging gold nanoparticles as selective Raman reporters and a silver nanoparticle-modified porous silicon microarray (Ag-pSi) as the SERS substrate. In the initial phase, experiments focused on optimizing physical parameters and conditions, leading to the successful construction of an assay capable of detecting bacterial cells, specifically *E. coli*, *S. aureus*, and *B. cereus*, within the concentration range of log₁ to log₅ CFU/mL, with detection limits of 6, 6, and 5 CFU/mL, respectively. The assay's selectivity was validated through experiments involving common interfering pathogens. Subsequently, the developed assay was applied to detect specified bacterial cells in various milk qualities and presented recovery values ranging from 89% to 107%. Notably, the overall assay timing was maintained under 90 minutes. These findings underscore the efficacy of the multiplex detection approach for bacterial pathogens in diverse samples, representing a significant advancement toward the realization of a portable SERS biosensor.

Session 59

Theatre 7

Are MIR calibration datasets representative enough of cow milk MIR spectra real population?

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Milk mid infrared (MIR) spectra have been used to develop models for predicting various traits, like milk fatty acids (FA). To ensure a good quality of the obtained MIR predictions, the collection of samples used to create the calibration set is supposed to be representative of the studied cow population. Yet this statement is not verified. This check is crucial in Holicow Interreg NWE Project as MIR spectra coming from different countries are aggregated and lack of spectral representativity would induce extrapolation. So, this study proposes a method driven by real data to solve this issue. Two databases were used. The first one included 163,657 spectra representing the cow population (DB1), the second contained 2,000 FA references and their MIR spectra (DB2). To assess if DB2 is spectrally representative of DB1, spectra were first decomposed into 3 principal components (PC). Normal distribution of data along each PC axis allows a simple comparison: is the sphere defined by DB2 records encapsulating the one defined by DB1 records? However, the actual distribution of data requires to prefer convex hulls over spheres. Volume of DB2 represented 7.43% of the DB1 volume without considering the density of DB1 records in the studied population. However, DB2 covered 99.44% of the DB1 density weighted records, the rest being extreme cases. It indicates that FA equations derived from DB2 should be suited for the vast majority of the 41 million of milk spectra included in the Holicow DB.

Valorization of bioactive components in milk: exploring environmental and animal factors affecting the variability of milk minerals in dairy cattle

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The objective of this study was to investigate the variability Ca, Mg, P, S, Na, K, and Cl in bovine milk to enhance and optimize their profiles for high-quality dairy products that meet consumers' nutritional needs. Milk samples were obtained from 1,080 cows reared in 54 herds located in the north and central regions of Italy. Cows were sampled once during evening milking. The elements were analyzed using the wavelength dispersive x-ray fluorescence technique. Minerals analysis employed a mixed model including animal [parity, days in milk (DIM), and daily milk yield] and environment [herd, season, altitude, temperature humidity index (THI)] -related factors. Overall, DIM affected all the investigated minerals ($P < 0.001$), with an increasing pattern for all throughout lactation period, except for K. Milk from multiparous cows had a lower quantity of K and P ($P < 0.001$) and higher levels of Cl and Na respect to primiparous ($P < 0.001$). Season notably impacted most of the minerals studied, with spring milk displaying the lowest content of Ca, K, S, Na, whereas milk produced in summer had the lowest Mg content. Altitude and THI were marginally associated to the variability of minerals. Milk samples collected in herds located in the mountains had higher levels of K ($P < 0.01$) compared to those collected from the plains and hills. In this context, THI did not play a significant role, indicating minimal changes in milk mineral composition under heat stress conditions.

Session 59

Theatre 9

Bulk milk quality traits differ according to the ClassyFarm welfare assessment score

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The Italian on-farm welfare assessment protocol, named ClassyFarm, evaluates the risk associated with farm housing and management conditions (resource-based indicators) united to the effects of the farming system on the animals (animal-based indicators). It returns an overall welfare score expressed into percentage from 0 (poor) to 100 (excellent). This study aimed to investigate the effect of the farm welfare score on bulk milk quality traits in dairy cow farms, differentiating between loose housing (LH) and tie-stall (TS) systems. Welfare protocol data of 194 farms (89 TS and 105 LH) located in Northern Italy were combined with their milk analyses retrieved from the milk payment systems archive of 2022 ($n=6,971$) for a retrospective study. A mixed linear model was used for variance analysis considering welfare score classes (insufficient, good, excellent), housing (LH and TS), month, and interaction between housing and welfare as fixed effects. Herd-season was the random effect. For TS, 90% of farms were classified as good, 4% as excellent, 6% as insufficient, while for LH, the corresponding percentages were 75%, 20%, and 5%, respectively. Welfare class and its interaction with housing type affected fat, protein, casein, lactose content as well as somatic cell score (SCS), total bacterial count (TBC) and fat-to-protein ratio. Farms in the worst welfare class exhibited the highest TBC and SCS and the lowest lactose content. The impact of rearing conditions on milk quality remains a complex matter because of the interplay of numerous factors and lack of knowledge regarding cause-effect relation.

Evaluation of MIR predicted milk components as biomarkers of heat stress in dairy cows using a residual approach

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Heat stress detection in dairy cows is still a challenge. For a diversity of phenotypes potentially linked with heat stress numerous prediction equations from mid-infrared (MIR) spectra of milk exist and are available in routine. However, confusion between heat stress and other factors affecting milk composition, like days in milk (DIM) or feeding make inference difficult. On this basis, the objective of this study was to use an alternative strategy to identify interesting biomarkers among a subset of milk components predicted by MIR. Rather than attempting to simultaneously account for all effects, this study employed a random regression model on DIM to first estimate residuals. Those were then used to evaluate the relative reaction amplitude of the traits with the temperature and humidity index (THI) and to estimate their common reaction to THI with performance traits. To perform this, 611,063 records from 97,042 Walloon primiparous Holstein cows, obtained from 2015 to 2022, were used. By focusing on residuals, unexpected variations with the THI were removed and clearer thresholds at which the molecules start to react with it were obtained compared to analysing the direct phenotypes. Milk components presenting the highest relative residual variation with the THI are the protein percentage, the milk concentration of polyunsaturated fatty acids (PUFA) and the milk concentration of magnesium. In addition, the reactions of those components to THI were favourably or neutrally correlated with the decrease of performance traits in the same conditions. In the next step detected milk components will enter further studies to develop comprehensive MIR-based heat stress indicators.

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Theatre 11

Analysis of antioxidant activity in bovine milk to valorize its nutraceutical properties

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The antioxidant activity of milk modulates the inhibition of the oxidation of nutrients, thus slowing down oxidative chain reactions. The antioxidant ability of milk is mainly correlated to casein, whey proteins, vitamin E and enzymatic antioxidants including superoxide dismutase, catalase and glutathione peroxidase. The variability of antioxidants in milk, both as quantity and as type, is influenced by several factors, some of which are environmental (season, diet, management strategies), while others are animal-related factors (species, breed, lactation stage). The purpose of this study was to analyze environmental (herd, season) and animal [parity, days in milk (DIM), daily milk yield] factors related to the variability of the antioxidant activity of individual samples of bovine milk. A total of 1,059 milk samples were collected once during evening milking. Cows belonged to 53 herds (20 cows/herd) located in northern Italy. Total antioxidant activity was assessed by the ferric reducing antioxidant power (FRAP; expressed as μM eq. ascorbic acid) and the 1,2-diphenyl-2-picrylhydrazyl (DPPH; expressed as % inhibition) methods. The results showed that FRAP was influenced by several factors such as DIM ($P < 0.05$), parity ($P < 0.05$), daily milk yield ($P < 0.01$) and season ($P < 0.001$). On the contrary, DPPH was not affected by any of the fixed effects included in the model, except for DIM factor. However, 70% of its variance was explained by the random effect of herd, meaning that this trait is probably influenced to a greater extent by environmental factors. Additional studies are needed to further investigate the environmental and genetic variation of these phenotypes at the individual animal level.

Evaluation of Shiga toxin-producing *Escherichia Coli* (STEC) in Traditional Dairy farms in Trentino
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Shiga toxin-producing *Escherichia coli* (STEC) are foodborne pathogens that pose a global public health and food safety challenge. The aims were to evaluate the presence of STEC in Trentino province (North Eastern Italy) dairy farms and possible correlations between STEC presence and milk quality parameters. The pathogen's presence was screened monthly in 15 farms delivering the milk to the local dairy factory for raw milk cheese production. We collected bulk milk samples, milking filters and environmental samples by overboots swabs. Milk samples were homogenized, decimally diluted in sterile peptone water and plated for the research of total bacterial count, lactococci, lactobacilli, coliforms, hemolytic streptococci and Prototheca spp. Milking filters and environmental samples were enriched with selective different antibiotics. All filters and environmental samples were tested by multiplex-PCR for the detection of the genes. Positive enrichments were subcultured on SMAC; positive colonies were isolated as putative STEC and tested by multiplex-PCR analysis. Preliminary results showed no positive correlation between the presence of Shiga toxin-producing genes and coliform count (mean 1.48 log UFC/mL, range 0.97 to 2.63 log UFC/mL). Moreover, despite having a positive presence in filters, many of the environmental samples were negative. Further investigations are scheduled to perform Whole Genome Sequencing (WGS) on more than 300 isolates. This study was carried out within the ONFoods2 and received funding from the European Union Next-GenerationEU (PNRR-MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.3–D.D. 1550 11/10/2022, PE00000003).

Identification of apathogenic bacteria as a reservoir for antibiotic resistance genes in bulk tank milk samples and in-line milk filters

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The intramammary use of antimicrobial agents in dairy farming is a controversial issue in the context of increasing antimicrobial resistance (AMR) in dairy farming. The widespread generalised use of antibiotic drying-off agents can lead to selection pressure on the bacteria, which promotes resistance. Particular attention is being paid here to the occurrence of methicillin-resistant *Staphylococcus (S.) aureus* (MRSA). The methicillin resistance of MRSA is mainly encoded by the *mecA* gene, which is located on a mobile element called SCC_{mec} that enables transmission to other *Staphylococcus* species. Recent studies suggest that the element has been transferred from coagulase-negative staphylococci (CoNS) to *S. aureus*. One way to test for the presence of certain antibiotic-resistant genes on dairy farms is to analyse tank and raw milk as well as the milk filters used. Studies have shown that the microbiome and AMR-determinants detected on milk filters can be considered representative of the microbiome and resistome of milk and its production environment. Milk filter and tank milk samples as well as rinse samples were taken. Samples were then cultivated on culture media and the presence of *mecA* was then analysed by PCR. The *mecA* gene was detected in the tank milk, in the rinse sample and in the five milk filters analysed. In addition, *S. sciuri* was identified as a reservoir for *mecA* in the rinse sample and in the four milk filters and *S. aureus* in one milk filter. The detection of *mecA* in analysed milk filter samples does not initially allow any direct conclusions to be drawn about the resistance situation of important udder pathogens, as the CoNS species *S. sciuri* does not necessarily have to originate from the udder. This is to be determined retrospectively using the individual animal milk samples, which were taken in parallel with the milk filter sampling. Regardless of the source of entry, *S. sciuri*-isolates in combination with the *mecA* gene were identified not only in the milk filter but also in the tank milk itself, which makes the hygiene measures on the farm under investigation even more important.

Development of a sandwich ELISA for the detection of bovine A1 beta-casein

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The beta-casein is one of major milk protein and classified mainly into A1 and A2 types. The A1 beta-casein harbours the histidine residue at the 67th amino acid position, while the A2 harbours the proline at the position. The amino acid substitution is derived from a single nucleotide substitution of A to C in the exon 7 of the bovine beta-casein 2 gene. Milk produced by cows homozygous for the A2 allele is known as A2 milk. A2 milk is thought to be less likely to cause stomach aches in people who experience lactose indigestion or intolerance. Some Japanese dairy farms are focusing on producing A2 milk. To produce A2 milk, the farms have to establish a herd consisting of solely of A2 homozygous cows. However, even after establishing such herd, there are risks to contaminate A1 milk into A2 milk. To solve this problem, we developed an ELISA method to detect A1 in the milk to be consumed as A2 milk. This ELISA method used an anti-bovine A1 beta-casein we developed in this study. The ELISA system could detect the A1 beta-casein when the concentration of it was more than 2% in A2 milk both before and after ultra-high temperature processing, which would allow us to perform the quality control of A2 milk.

Characterization of Dual-Purpose Blue milk compared to Holstein milk based on mid-infrared spectrometry

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There is a growing interest in developing local breed-derived products as cheese. However, often, little is known about the milk properties of local breeds. The objective of this study was to compare milk spectral variability of the Holstein (HOL) and the local Dual-Purpose Blue (DPB) cattle breeds. For this purpose, we selected representative milk mid-infrared (MIR) spectra from each breed. First, localization index were computed using scores of spectra projected on the first three dimensions of a principal component analysis (PCA) based on one million Walloon (Southern Belgium) spectra. A total of 165,507 HOL and 53,561 DPB spectra were selected. We then made a PCA on the selected spectra and projected supplementary variables, namely MIR predictions of milk composition and processability to dairy products, on the PCA. On the first two dimensions of the PCA, we detected a spectral variability of DPB that was not covered by HOL. We further had a look to supplementary variables with \cos^2 higher than 0.4 in that PCA region and found that these DPB spectra were characterized by higher K20 (longer time to obtain a firm curd of 20 mm), lower casein content, protein percentage and sodium content than HOL. This would mean that DPB milk related to these spectra were not suitable for cheese processing. However, there were also DPB spectra characterized by high fresh and dry cheese yield in another region of the PCA. As a previous study found that K20 properties, casein and protein percentages of milk are highly heritable in DPB, breeders interested in good cheese making properties should breed cows with spectra found in the PCA region related to high fresh and dry cheese yield.

Does the halloumi cheese production process change the fatty acid profile of milk used?

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This study tested the fatty acid (FA) profile of milk and halloumi cheese, produced by feeding animals with or without olive cake. The hypothesis to test was whether the halloumi cheese process affects the FA profile of milk by changing or not the unsaturation of lipids. For this purpose, 24 mid-lactated cows were fed with 2 isonitrogenous and isoenergetic diets, named the control and the olive cake (OC) diet, where part of the forages were replaced with ensiled OC as 10% of dry matter according to a 2 × 2 crossover design with two 28-d experimental periods. At the end of the second experimental period, bulk milk obtained from cows of each treatment was converted into 7 different halloumi cheeses (14 in total). Dietary supplementation with ensiled OC modified the FA profile of milk, and thereby the fat of halloumi cheese produced. The concentration of de novo synthesized FA, saturated FA, and the atherogenic index was decreased, while long-chain and monounsaturated FA content was increased in milk, and this was transferred to halloumi cheese after the process. Individual saturated FA like stearic acid was elevated, whereas among individual monounsaturated FA, increments of oleic acid (C18:1 cis-9) and the sum of C18:1 trans-10 and trans-11 acids were demonstrated in milk and halloumi cheese produced. By feeding cows with OC, the concentration of CLA cis-9, trans-11 was increased in both milk and halloumi cheese fat. Overall, the halloumi cheese production process did not affect the FA profile of lipids and therefore the inclusion of any oily feed, such as ensiled OC, can improve, beneficially for human health, the lipid profile of halloumi cheese produced.

Session 59

Poster 17

Public awareness of iodine in human health and diet as assessed by cross cultural survey

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According to the World Health Organization, iodine deficiency can lead to brain damage. Animal-origin foods, particularly dairy, are significant iodine sources. This international survey evaluates public awareness of iodine's significance for human health and dietary sources. Sociodemographic details covered country, gender, age, education, and employment. Likert scale questions (from 1=not at all to 7=very much) gauged perceptions on the impact of iodine on health and on the associations between iodine and milk, fish, seafood, meat, vegetables, and fruit. The survey garnered 4,704 responses from 16 countries. Data were analyzed through a multiple regression model, accounting for country, gender, age, education level, and employment status. All countries recognized the influence of dietary iodine on human health. Respondents from New Zealand, Greece, and Italy exhibited the highest awareness of iodine's impact on health (5.91, 5.64, 5.64 respectively). The recognition of milk as iodine-rich food was low, especially in Northern and Western Europe (2.96). Results highlighted strong awareness of fish and seafood as iodine sources. Northern America scored highest in perceiving meat as a dietary iodine source (4.66). Eastern, Southern, and Western Asia showed higher awareness of vegetables, fruits, and cereals as dietary iodine sources compared to Northern and Western Europe. Results demonstrated a low knowledge about iodine dietary sources other than fish. To enhance public awareness, authorities should encourage the use of informative packaging labels on iodine-rich foods. This work was financed by Bando 2022 PNRRProt. P20228RP2Z

Assessing solutions for resilient dairy farming in Europe

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The European dairy sector faces a set of challenges. Project Resilience for Dairy (R4D) deals with these challenges (<https://resilience4dairy.eu>) by developing a Thematic Network on 100 resilient dairy farms in 15 EU-countries. Our research is based on assessment of solutions (practices, techniques, tools) that contribute to future resilient dairy farming. Firstly, farmer needs were collected during 2021 by a survey filled in by 535 stakeholders the participating countries. Next, outcomes were discussed in national dairy AKIS (NDA) meetings and in European expert workshops, resulting in the formulation of 190 possible solutions in the resilience areas: socioeconomics, technical efficiency, and environment/animal welfare/health. The solutions were evaluated using a developed assessment scheme (Likert scale: scores 1 to 5). A total of 66 expert assessors, selected by the participating organizations did perform 3329 assessments with focus on resilience. Additionally, the readiness and attractiveness of the solutions were evaluated in NDA meetings in 2022/23. Each NDA group was asked to select the 20 solutions with highest attractiveness, resilience, and readiness. There was variation in focus over Europe, especially in the NDA groups, and, also, expert' and stakeholder' opinions appeared to differ. Technical efficiency remains a leading strategy at farm level. Improved labour organisation, strategic hoof trimming, early detection of diseases, hospital pens, technology for heat stress management, various aspects of calf rearing, and agro forestry were much mentioned topics of interest, while communication with society, renewable energy production and use of farm economic indicators, among other, were specifically emphasised by farmers and stakeholders. Choices of solutions were affected by facilitation, choice of farmer groups, and societal unrest during the study period.

Session 60

Theatre 2

WELL-E: Enhancing Animal Welfare & Longevity through Artificial Intelligence and Internet of Things Dedicated to Dairy Farming

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The WELL-E initiative is constituted as a Digital Living Lab centered on animal and end user needs. Scientific evidence shows how animal welfare and cow longevity go hand in hand, driving the industry's interest in cow welfare, as improving longevity is considered an answer to issues of economic, social, and environmental sustainability in dairy farming. Our approach is based on the use of IoT, computer vision, and machine learning to enhance our ability to detect and monitor changes in welfare and longevity before the onset of visible outcomes and to generate predictions to aid in on-farm decision-making, ensuring that efforts are focused on animals most likely to succeed in the long-term. Launched in 2023, our team has conducted pilot research and worked on building functional and resilient data collection infrastructures for implementation on commercial dairies in 2025. We also developed a framework for the study of animal behaviors and emotions, and a paradigm shift for both data annotation and analytics based on continuous and heterogeneous source of data. This research combining animal welfare and technological development, especially on aspects regarding the implementation of integrated virtual ecosystems aiming to enable real-time monitoring and decision-making, will help farmers anchor their decisions in a solid foundation of data and give new keys to the whole livestock value chain for ensuring its sustainability.

Viable future dairy farming systems: a comprehensive nutritional approach to minimize methane emission and phosphorus and nitrogen excretion

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Dairy farming contributes to environmental pollution through methane emission and excess phosphorus (P) and nitrogen (N) on farms. We outline nutritional approaches to lower P and N excretion and methane emission in dairy cattle, focusing on the Dutch dairy sector as a case. Phosphorus excretion in the Dutch dairy sector significantly decreased, largely due to a voluntary feed covenant limiting concentrate P content. Recent evidence challenges current dietary P recommendations, suggesting lower levels to effectively reduce P excretion whilst enhancing cow health. While N excretion in the Dutch dairy sector remained rather stable over the past decade, lowering dietary N content shows promise in reducing N excretion, albeit not readily adopted due to concerns about perceived milk production losses. Addressing this challenge requires recognizing the role of absorbed energy and essential amino acid profile in improving N efficiency at cow level. Methane emission of Dutch dairy cattle increased in the past decade. Evaluating methane mitigation strategies is crucial, as recent meta-analyses highlight the ineffectiveness of various methods. High forage quality is key in decreasing methane emissions, with future support from effective anti-methanogenic feed additives. Focus on reduction of one pollutant may have implications for emissions of other pollutants. For example, several measures to reduce N losses at farm level, including reduced maize cultivation, increased methane emissions. An integral approach is imperative to further decrease P and N excretion as well as methane emission in dairy cattle.

Session 60

Theatre 4

Breeding for resilience in the Netherlands and Flanders

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Dairy cows could face several environmental disturbances during a lactation like weather conditions or changes in roughage quality, resulting in a reduced functioning. Cows which are minimally affected by disturbances and/or quickly recover are the preferred cows. Reduced functioning of dairy cows is measured as the difference in daily milk yield and the expected milk yield for that day. This is called deviation. The expected daily milk yield is estimated with polynomial quantile regression based on all milkings of a cow during a lactation. Based on the deviations, two resilience traits are calculated: stability and recovery. Stability is the natural logarithm of the variance (LnVar) from all deviations during a lactation, recovery is the autocorrelation (Rauto) between all deviations during a lactation. A lower LnVar indicates less affection by disturbances, a lower Rauto indicates quicker recovery. Breeding values are estimated for stability and recovery for lactation 1, lactation 2 and lactation 3 and later (3+). Heritability (h^2) is 0.09, 0.06 and 0.09 for stability and 0.07, 0.04 and 0.04 for recovery for respectively lactation 1, 2 and 3+. Genetic correlations between different parities for stability ranges from 0.91 to 0.98 and for recovery it ranges from 0.84 to 1.00. Overall breeding values for stability and recovery are calculated based on traits in lactation 1, 2 and 3+. An overall index for resilience is calculated based on the overall breeding values for stability and recovery. Breeding on resilience results in cows that are less affected by environmental disturbances and recover more quickly. The overall index and the two overall breeding values are the traits that are published in the Netherlands and Flanders since April 2024.

If you were a cow?

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The aim of this study was to clarify the concept of eco-citizen dairy farming developed in the Coccinelle project, from the mental representations of visitors to a public event held near Clermont-Ferrand (300,000 inhabitants), in response to the question: If you were a cow, what would you prefer? Forty groups of 1 to 5 people answered a photo-based quiz. For each of 12 themes proposed (e.g. What do you prefer to eat in winter?) participants chose between 3 answers (e.g. hay, maize silage or wrapped bales) illustrated by a photo and a short explanatory text. The photo illustrating the situation judged as POSitive was placed in a specific box. The two remaining photos were placed in the NEUtral or NEGative boxes. An anonymous questionnaire characterised each participant (gender, age, residence), their frequency of contact with farmers and their sensitivity to animal welfare. The data collected was processed by factorial correspondence analysis, then by cluster classification. Axis 1 of the analyse represents 73% of the variability in responses and opposes POS practices with NEU or NEG. Cluster 1 groups together 77% of situations judged POS, 19% of NEU and 4% of NEG. In contrast, cluster 3 groups 63% of NEG situations, 30% of NEU and 7% of POS. For participants, the ideal would be to be a cow of a rustic breed with horns, mated to a dairy bull, rearing its calf, housed in a free stall with straw and a rotating brush for scratching. Fed hay in winter, she benefits from a mobile milking parlour and shaded pastures surrounded by electric wires in summer. She has positive contact with the farmer, who also looks after her health. As this ideal is far from reality on the majority of French farms, citizens and farmers need to discuss how to reduce the gap between them.

Session 60

Theatre 6

Mitigation of environmental impacts in dairy farming: potential synergies and trade-offs

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Livestock farming frequently faces strong criticism for its greenhouse gas emissions (GHGE) and impacts of nutrient surpluses like terrestrial acidification and eutrophication, but also for use of potential human food resources. Based on the results of a Life Cycle Assessment (LCA) of 89 dairy farms in grassland dominated areas of Austria, our research aims to explore drivers of GHGE per kg ECM and to explain differences between farms as well as potential trade-offs and synergies for addressing multiple environmental impact categories. Results of the LCA vary strongly between single farms, thus highlighting existing potentials for improvement. GHGE per kg ECM are predominantly determined by milk yield and cumulative fossil energy use. High milk yields are associated with low land use but also increased utilization of potentially human edible feedstuffs and nutrient losses per hectare. Furthermore, results show a clear association with site conditions and thus call for further in-depth analysis to ensure the adequacy of potential mitigation measures, particularly for less favourable areas. Acknowledgements: The authors would like to thank SalzburgMilch GmbH for the cooperation and partial funding of the research.

Regional feed and energy centre

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Cooperation between dairy and arable farmers can help to optimize sustainability on regional level instead of on farm level. The example of a regional feed centre will be illustrated. A feed centre buys crops from dairy farmers and arable farmers, processes these into total rations and sells them to a number of dairy farmers, to be delivered at the feeding fence. A study will be presented that aims to assess the sustainability of a regional feed centre in the Netherlands on the basis of economics, traffic movements, energy consumption and landscape. Thanks to a feed centre, the cost price on dairy farms can decrease by € 1.80 to € 3.30 per 100 kg milk in the long term, depending on a regime of grazing or non-grazing and the quantity of land available. Even though the number of traffic movements increases when using a feed centre, the total energy consumption decreases. The farm surroundings improve as on-farm feed storage facilities will disappear and a greater diversity of crops is grown. Another advantage is that grassland management may improve because reliable data about the quality and quantity of grass will become available. Additionally, a study about an Energy Cooperative will be presented. The study concerns the possibility of teamwise mono-manure fermentation of 26 dairy farms and the effect on the reduction of ammonia and greenhouse gas emissions was examined. If slurry is removed daily from the cattle barn, fermented after a short storage time and the digestate is stripped, an ammonia emission reduction of 46% can be achieved over the entire chain, as well as a greenhouse gas emission reduction of 78% (enteric emissions excluded). However, this is highly dependent on the chosen low-emission housing system and how quickly liquid manure is removed from the barn and further processed. Furthermore, assuming that RENURE fertilizers (a manure mineral concentrate) may be used as fertilizer, a saving of 160 tons artificial fertilizer per year can be achieved (equal to approximately € 700,000) and with the central fermentation of manure and upgrading to green gas an equivalent of 1,864 tons of CO₂ emissions can be prevented.

Session 60

Theatre 8

Simulating environmental sustainability of Dutch dairy farms towards 2030

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Dairy production systems are causing severe pressure on various sustainability aspects, such as global warming, eutrophication, acidification and biodiversity loss. The objective of this study was to evaluate effects of changes in farm structure towards 2030 following farm expansion, productivity increases, and implications of Dutch policy measures on environmental sustainability performance of various types of Dutch dairy farms. First, 6 types of dairy farms were described varying in soil type (clay, sand, peat), herd size, intensity (milk per ha), grazing, and conventional/organic farming. For each farm, farm structure and performance were based on data representative of comparable Dutch dairy farms in 2022. The farms were modeled in DairyWise, an empirical farm-scale model to simulate technical, environmental, and financial processes on dairy farms. Next, for the 2030 situation assumptions were made with regard to: expansion of herd size and farmland based on other farms closing and phosphate rights; increases in milk yield and forage yields based on historical trends, combined with a higher precipitation deficit due to climate change; a higher groundwater table in peat soils; mandatory crop rotation and catch crops on sandy soils; changes in manure allocation and mineral fertilizer use due to loss of derogation, buffer strips, and limitations to total nitrogen use; and a decrease in crude protein level of the feed ration to 160 g/kg DM on average across all farms. Assumed changes were simulated for each of the 6 farms in DairyWise. Results showed a strong reduction in soil nitrogen surplus and ammonia emissions on the conventional dairy farms, but also a strong increase in the amount of manure exported from farms. Greenhouse gas emissions reduced, and grazing time increased on all farms. We concluded that the loss of derogation, buffer strips, stricter nitrogen standards, and reduced crude protein level had large implications for on-farm nitrogen emissions. In a next step, effects of alternative development pathways for the 6 dairy farms will be explored.

Advancing precision extensive livestock farming through AI-based species recognition from camera trap recordings

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Interactions among domestic and wild ungulates in extensive livestock farming systems can profoundly impact their welfare, ecosystem dynamics, biodiversity, and the sustainability of pastoral and forestry practices. Automatic photo-video recording systems like camera traps offer continuous observation of animals without interference and at minimal cost. However, post-processing requires manually assigning species from videos or photos for database construction, which is very time-consuming. In this study, we trained an AI algorithm to recognize species from camera trap images, facilitating automatic database construction. We strategically placed 18 camera traps in grazing areas using a random systematic sampling approach. A dataset of 1320 images was annotated with species identification (cows, red deer, roe deer, and wild boar) using the Roboflow platform. Data augmentation to 2382 enhanced dataset diversity, addressing overfitting and biases during training. Ultralytics' YOLOv8 object detection algorithm was run on a laptop equipped with an NVIDIA® GeForce RTX™ 3060. Performance metrics, including Accuracy (A, mAP@50-95), Precision (P), and Recall (R), were evaluated based on Intersection over Union between predicted and ground truth bounding boxes. Results showed a mAP@50-95 of 0.696 for all classes, with P at 0.847 and R at 0.743. The highest metrics were observed for cows and wild boars, with mAP@50-95 values of 0.722 and 0.805 respectively. P scores were 0.968 for cows and 0.743 for wild boars, while R values stood at 0.847 and 0.971, respectively. Lower values were for deer and roe deer. Overall, the model demonstrated 91% correct assignment for cows, 86% for deer, 47% for roe deer, and 99% for wild boar. Implementation of object detection models through AI for identifying species from camera trap images offers a promising avenue for extensive precision livestock farming, facilitating efficient monitoring of wild and farmed animal interactions and biodiversity conservation.

Session 60

Theatre 10

What are the limits for agricultural collectives under Geographical indications (GI) to take action in transitions? Six case studies in French cheese sector from the ADAOPT project

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Collective support in livestock sectors is a major issue to face sustainable transitions (Lema et al, 2021, Scotti et al, 2023), but it raises complex questions about practical implementation in sectors under GI (Dernat et al, 2022). ADAOPT project wants to mobilise the whole GI collective in French cheese sectors. Six pilot GIs are aiming to design and implement their own collective strategy to face transitions. The mobilisation methods used range on two metrics: i) from a large organisation scale covering the whole GI's territory to small local working group, ii) from a focal on technical issues to a global understanding. A transversal analysis was set up to follow the GIs. This project shows the difficulty of mobilising GIs actors, despite the diversity of participatory methods used. Results show that several parameters inherent in GIs limits the support: The geographical extent of some of the GIs and the inherent heterogeneity of production systems and climatic conditions. The lack of clear governance (traditional top-down organisation), which usually delegates the investigation of a topic to a non-representative working group. Most of the levers for adaptation are related to production, which faces challenges in engaging in sector-wide debates due to political and socio-economic obstacles. The dependence on processors, who can restrict the choices made by the sector and farmers. The overlap between the players providing support in the territories, generating an over-solicitation on similar subjects but pursuing different goals. This work highlights the need to develop support methods that will make it easier for collectives under GI to act.

Effect of feeding a garlic-citrus extract on milk production of dairy cows and sensory perception of milk
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Several studies have shown the anti-methanogenic effect of feeding a garlic-citrus extract supplement (GCE) to lactating cows. However, there is limited data on the effect of GCE supplementation on the sensory properties of milk. This study evaluated the effect of GCE supplementation in dairy cows on milk production and sensory perception of milk. Twenty-one Holstein Friesian cows were randomly assigned either to a diet without (control, n = 11) or with 30 g of GCE/cow/d (GCE, n = 10). The study comprised 3 weeks of covariate period followed by 7 weeks during which milk yield and composition were measured. Total urinary N (UN), urinary urea N (UUN) and NH₃ emissions were estimated using milk urea N (MUN). In week 7, the sensory attributes related to colour, aroma, texture and flavour of pasteurized milk samples were evaluated by 34 untrained consumers. Data were statistically analyzed using a linear mixed model in SPSS. Supplementation with GCE did not affect milk yield or composition parameters (fat%, protein% and lactose%). GCE cows had lower somatic cell count (SCC; -25%) and MUN levels (-14%) compared to the control. Consequently, GCE cows exhibited lower predicted UN (-12%), UUN (-20%), and NH₃ emissions (-10 to -12%). The sensory properties and consumer acceptance of milk were not affected by GCE supplementation. Overall, these results indicate that GCE supplementation had no effect on milk production but reduced the SCC and MUN levels, suggesting potential positive impacts on udder health and environmental mitigation of N emissions. Moreover, feeding GCE did not influence the sensory perception of milk.

Effects of thermally processed linseed on meat quality traits of fattening Holstein calves

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Self-sufficiency of beef production in Greece accounts for about 25%, and domestic demands are covered mainly by imports. Increasing domestic production is challenging, and using calves from dairy herds could be an option. The objective was to investigate whether designated feeding of male Holstein calves could produce carcasses with high-quality characteristics. Male calves originated from two farms with 350±70.71 cows and 212±190.92 heifers. In each farm, 40 calves were randomly selected and equally allocated into two groups (Control C, n=20 and Treatment T, n=20), further separated into four subgroups each. During the experimental period, lasting 90 days, both groups received a total mixed ration composed primarily of maize silage, straw, ground corn and soybean meal. The experimental diet included 4% extruded linseed, replacing the equivalent amount of ground corn. Feeding and water were provided ad libitum. Feed residuals and body weight measurements are performed monthly. Average age and body weight at slaughter were 17±1.0 months and 555±0.70Kg, respectively. Produced meat is assessed by collecting individual meat samples (13th rib) for quality assessment and physicochemical analyses. A comparative analysis is performed to test for differences in growth potential and meat quality traits of produced meat. The project is expected to support the transitioning of the dairy cow sector into a production system that supports both milk and meat production and, hence, enhances economic viability. This work is funded by "BlackWhite" project "Measure 16 'Cooperation'" (Project code M16SYN2-00024).

REGENEMILK: Innovative strategies and techniques for monitoring and treatment of mastitis in local and cosmopolitan breeds

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The Regenemilk project aims to determine the effectiveness of a biological product, the platelet-rich plasma (PRP) which is a preparation based on platelet factors (e.g., chemokines, cytokines and active metabolites) extracted from blood, as a possible alternative to conventional antibiotic therapy for the mastitis in dairy cattle. To identify the clinical cases, udder health of a total of 400 cows (200 Holstein Friesian and 200 Reggiana) will be monitored for 18 months and somatic cell traits and bacteriological characterization of milk will be provided. Cows with mastitis will be divided in two groups depending on the treatment (PRP vs conventional therapy). The udder health status of the cows will be assessed via several milk traits related to the health status (i.e., somatic cell count, antibiogram pattern of bacteria) and milk quality (i.e., composition, cheese-making properties). Regenemilk will also extend its focus to assess the influence of the bovine breed on ability to recover from a mastitis and to investigate the potential effect of inbreeding depression in cosmopolitan and local breeds. The validation of the use of this innovative treatment could pave the way for a reduction in antibiotic usage and economic losses in dairy cattle.

Session 61

Theatre 1

Effectiveness of a commercial Diatomaceous earth feed additive in mitigating transport stress in Sea Bass (*Dicentrarchus labrax*)

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The aim of the work is to assess the effects of Dorabrin® (78% of Diatomaceous earth) dietary supplementation in European Sea bass during transport. Two groups of fish (20.56±1.30g LW) were farmed in two aquarium of 30m³ (density of 35kg/m³), both fed with same commercial feed, one without (CON) and one with the supplementation (EXP) of 800g/ton of feed of Dorabrin®, the feeding trial was carried out for 9 days before transport. On the 10th day, both treatments (CON and EXP) were transferred into 6 tanks (2m³; 55kg/m³) (about 5200 animals for each tank) for the transport. Blood and skin mucus were collected from a subsample pool of 20 animals from each tank, randomly caught, in 3 different experimental time: 30 min. before of the handling (T0), 1h after the transfer but 20 min. before the trip (T1) and 30 min. after the transport of 3h (T2). After testing data normal distribution, an ANOVA was carried out, corrected for repeated measures and a Pearson correlation was performed. In both the experimental groups plasma cortisol increased significantly from T0 to T2 (P<0.01), with lower values in EXP compared to CON group at T2 (P<0.01). Glucose and lactate increased significantly (P<0.01) in both blood and skin mucus of both the experimental groups, showing, at T2 (P<0.01), lower values in EXP one. Although, stress indices in EXP increased at T2, they remained significantly lower than in CON. Plasma and skin mucus cortisol showed to be highly correlated (R₂ = 0.76, P<0.01). Dorabrin® proved to be a feasible product for mitigating the effects of transport stress. The positive relationships between some indices in plasma and mucus opens to the possibility of determining stress in a simple, non-invasive and rapid way.

Diet digestibility and growth performance of African catfish fed diets with blends of poultry PAP

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With the general aim of increasing the efficiency and profitability of market-ready diets tailored for recirculating aquaculture system (RAS), this study evaluated diet digestibility and growth performance of African catfish (*Clarias gariepinus*) fed four practical diets (CP: 44% DM; EE: 12% DM; GE: 19 MJ kg⁻¹) containing different rates of fishmeal (FM) and blends of poultry processed animal proteins (PAP) meals, i.e. poultry by-product meal, hydrolysed feather meal and poultry dry-blood meal. A total of 600 fish (116 ± 16 g) were randomly distributed into 12 RAS units (3 tanks × treatment) and fed during 84 days with diets based on different levels of FM (from 9 to 1%) and increasing levels of PAP (from 23 to 39%), that is 9FM-23PAP, 5FM-28PAP, 9FM-31PAP, 1FM-39PAP. Fish fed diet 1FM-39PAP showed the lowest protein (-3%) and lipid (-2%) digestibility coefficients and the worst growth performance in terms of final body weight (-18%), total length (-7%), condition factor (-4%), feed intake (-18%), specific growth rate (-14%) and feed conversion ratio (+21%) compared to the other diets (P<0.05). Fish fed intermediate diet 5FM-28PAP reported the highest survival rate (97%) compared to other diets (P<0.001). Overall, also from an economic perspective, high PAP inclusion (39%) associated to very low FM (1%) worsened diet utilization and fish performance, whereas a viable dietary formulation for RAS farming of African catfish should include a more balanced and intermediate levels of the two protein sources.

Session 61

Theatre 3

Feed and carbon footprint in aquaculture – an industry perspective

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Aquaculture plays a major role in providing food for a growing world population while addressing challenges such as climate change and social impact. Feed is a major contributor to the total greenhouse gas emissions of farmed fish and shrimps and the feed industry can therefore take a leading role in lowering aquaculture carbon footprint. This requires adoption of robust and aligned methodologies to calculate impacts, full traceability in order to know where feed ingredients are produced or cultivated, availability of more primary data, consideration of other environmental impacts and of the full life cycle, and finally to deal with highly volatile prices of the raw materials. Sustainability is not a straightforward journey, and there are many challenges and dilemmas to be addressed together with opportunities not to be missed. This requires collaboration along the supply chain, including raw material suppliers, fish farmers and feed manufacturers on the same journey, to provide sustainable seafood for a growing world population.

The sustainable “4 F” for rainbow trout (*Oncorhynchus mykiss*) feeding: Feedstuffs from the Fish Farmer Farmhouse

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In rainbow trout production, feed sustainability, together animal welfare, quality of the final product and environmental impact are the main challenges to face for the operators. For the aim, a trial was carried out comparing two different feeds characterized by different dietary ingredient composition: D1 included fish meal and conventional feedstuffs and D2 with co-products deriving from fish processing as main protein and lipid source and vegetable feedstuffs coming from the farmhouse owned by the same fish farmer. 600 young rainbow trout (mean body weight=48±3g) were reared in 6 tanks resulting in 3 replicates per dietary treatment. The main physico-chemical water parameters were monitored in order to evaluate effects on the environment. At the end of the 8-week trial, the most important growth performances and quality of the fillet were determined in terms of proximate composition and fatty acid profile. Both the groups reached a suitable final mean body weight (D1=172±35.7g; D2=151.65±34.7g). The feed conversion showed a better rate in D2 (1.03±0.02) compared to D1 (1.3±0.03). Water quality exhibited an improvement of the main chemical parameters with a reduction of total ammonia nitrogen and nitrites in D2 (TAN=0.23 mg/l; NO₂-N=0.08 mg/l). No significant differences were observed in fish fillet quality and fatty acid profile. This study showed that dietary ingredient composition, as co-products deriving from animal processing, together with vegetable feedstuffs from the farmer farmhouse, favourably affected the sustainability of the trout production system.

Session 61

Theatre 5

Environmental impact of the inclusion of alternative raw materials in diets for gilthead seabream

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The study used Life Cycle Assessment (LCA) to analyse the effect on global warming potential (GWP) of the partial and total replacement of fishmeal with blends of alternative protein meals in diets for gilthead seabream. A total of 360 seabream (64 ± 1.3 g) were fed five diets (three tanks per diet): a control diet (FM100 with 590 g/kg fishmeal) or four alternative diets where fishmeal was replaced at 75% (FM25), 90% (FM10), and 100% (FM0) by protein blends consisting of Iberian pig meal (IPM, 237-328 g/kg) and vegetable protein meals (soybean, pea and sunflower meals) (373-478 g/kg). An additional fishmeal-free diet also contained 50 g/kg of microalgae *Isochrysis aff. galbana* (T-Iso) (FM0+). Final weight was higher (+48%; P<0.05) and feed conversion ratio lower (-29%; P<0.05) in fish fed FM100 and FM25 diets compared with FM0 diet, with intermediate values for FM10 and FM0+ diets. The LCA included the impact of aquafeed production, referred to 1 kg increase of fish. The GWP increased with fishmeal replacement, from 1.15 kg CO₂-eq in FM100 to 2.45 kg CO₂-eq in FM0 diets. In FM100 diet, fishmeal contributed to 47% of the total impact, followed by wheat meal (20%), and soybean oil (20%). In alternative diets, IPM was the major contributor to GWP (50% to 60%), followed by soybean meal (6% to 8). In FM0+ diet, T-Iso contributed to 14% of total GWP. Based on our preliminary results, the replacement of fishmeal at 75% with a blend of Iberian pig meal and vegetable proteins seems to represent the best trade-off, both in terms of growth and environmental performance.

Microplastics manifestation and their potential impact on freshwater fish species

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Microplastic (MPs) pollution in the aquatic environment is an emerging subject worldwide, but very few investigations have been reported in the riverine ecosystem. The current study assessed the occurrence and potentially damaging effects of microplastics on riverine fish population inhabited in the river Jhelum across the Punjab, Pakistan by involving three sites of the river (viz., Patan Machyana= JSI, Sakhira= JSII and Sawa Nankana= JSIII) because of high pollution loads and a sizable amount of floating plastic debris on the surface of water. Raman spectroscopy was used to confirm MPs polymer type and their distribution in water, sediments, and fish tissues (gut, gills, liver, and muscles). Scanning electron microscopy (SEM) was used to confirm the morphology of microplastics. MPs manifestation was observed in the order of JSIII > JSI > JSII and MPs morphotypes in water and sediments was fragments > fibers > sheets. Among the tissues, the gut showed the highest accumulation and fragments were more abundant at SI while fibers were highest at SII and SIII. Histopathological alterations in gut tissues were most obvious including villus degeneration and folding with mucosal inflammation, liver and muscle tissues showed mild to severe alterations, while gills were observed with severe physical changes due to direct accumulation of MPs. Raman spectra confirmed the chemical nature of MPs as polyethylene vinyl acetate (PEVA), polypropylene (PP), and polyethylene (PE). The results of this study showed that MP pollution poses an evolving risk to the fish of the riverine ecosystem and living under stress conditions.

Session 61

Theatre 7

Traceability and characterization of nanoplastics on novel feed/food sources: the case of microalgae

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Nanoplastics (NPs) pose a biological risk for human and animal health along the feed to food chain. In this work, we investigated by Dynamic Light Scattering (DLS) the effects of static, dynamic and ex vivo digestion on the size, polydispersity index (PdI) and Z-potential of PS (100 nm), PE (200 nm), PP (180 nm) and their mixture (MIX) after 24h contamination of *Chlorella vulgaris* (CV) and *Haematococcus pluvialis* (HP), microalgae widely used in the food/feed sector. In parallel, the modulation of total phenolic content (TPC), induced by NPs on CV and HP, before and after digestion and the pro-inflammatory effect on intestinal cells were evaluated. The data (mean±SEM) were analysed by one-way Anova (p-value<0.05). Before digestion, DLS confirmed the nominal size of PS (85.32±0.32nm) and PE (207.50±1.18nm) in CV medium, while PS+PE showed trend to agglomerate (267.20±4.84nm) with significant differences (p<0.05). Following in vitro static digestion, PS, PE and PS+PE increased in size (214.6±1.42nm; 500.97±4.58nm; 303.53±4.33nm, respectively). Before digestion, the MIX of PS+PE (202.10±6.97%) showed a significant higher TPC (p<0.05) compared to PS (137.86±2.44%) and PE (130.43±1.29%); results also confirmed after all digestion processes. Although, this work demonstrates for the first time the toxic potential of the NPs MIX also on intestinal cells, further evaluation will be fundamental to confirm this.

Effect of the low level dietary inclusion of queen bee larvae meal on the fatty acid profile of the fillets and livers of European seabass

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Amidst growing interest in using insects for animal feed, studies are investigating new species like *Apis mellifera* queen bee larvae (QBL). QBL, a byproduct of royal jelly production, offers promise for circular economy practices and possesses functional potential, due to royal jelly enrichment on their body left by the harvesting. This study aimed to assess the impact of dietary inclusion of queen bee larvae meal (QBLM) at 1%, 2%, or 3% (QBLM1, QBLM2, and QBLM3) on the fatty acid composition of *Dicentrarchus labrax* livers and fillets. Over 180 days, 120 fish (30 fish/treatment) were reared in 12 tanks. After euthanasia using clove oil, total lipids, fatty acid (FA) profiles, atherogenic index (IA), thrombogenic index (IT), and elongase and desaturase $\Delta 5 + \Delta 6$ (n-3) and (n-6) activities were determined, using one-way ANOVA for statistical analysis. Linoleic acid was significantly higher in the CTRL (no QBL inclusion) livers ($p = 0.0079$), whilst the linolenic acid was more abundant in QBLM3 group (4.17% total FAME) compared to the CTRL (3.78%) and the QBLM1 and QBLM2 (3.71 and 4.04%). Additionally, C20:3n-6 content was notably more present in the CTRL group ($p = 0.0032$). No differences were detected for IA, IT, or enzymatic activity in livers. In fillets, C16:0 peaked in QBLM1 ($p = 0.0168$), C20:0 was greater in CTRL than in the treatments, and C22:1n-9 was significantly higher in the QBLM3 (0.31% total FAME) than other groups. Contrarily to the liver, elongase activity was significantly lower in the QBLM1 fillets ($p = 0.0147$). Overall, QBLM up to 3% in European seabass diets did not notably enhance the fatty acid profile or the functional capacity of the fillets. However, it did not show any detrimental effects, particularly in the liver. Thus, further research is warranted for framing QBL reusing and its potential in animal feeds.

Session 62

Theatre 1

Effect of feeding a garlic-citrus extract on milk production and fatty acid profile of milk

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There is increasing evidence that feeding a garlic-citrus extract supplement (GCE) to lactating cows could improve milk production in addition to its anti-methanogenic effect. However, there is limited data on the effect of dietary GCE on milk fatty acids (FA). This study evaluated the effect of GCE supplementation in dairy cows on milk production, composition and FA profile. Thirty Holstein Friesian cows were randomly assigned either to a diet without (control, n=15) or with 1 g GCE/kg DMI (GCE, n=15). The study comprised 2 weeks of covariate period followed by 6 weeks during which milk yield (daily), composition and FA profile (weeks 2, 4 and 6) were measured. The milk FA% was determined using gas chromatographic analysis of FA methyl esters in milk. Data were statistically analyzed using a linear mixed model in SAS. Dietary GCE did not influence milk yield, protein% and lactose%, but increased fat% (4.62 vs 4.10%) compared to the control. This resulted in increased yields (kg/d) of fat (+0.2), total solids (+0.2), fat-corrected milk (FCM; +3.7) and energy-corrected milk (ECM; +2.8) in the GCE cows. The milk FA profile was similar for both groups except for higher C18:3 n-3 and lower C18:3 n-6, c9, t11-C18:2 and total conjugated linoleic acids in GCE cows. Moreover, similar nutritional indices (total saturated, monounsaturated, and polyunsaturated FA, n-6:n-3 FA, atherogenic and thrombogenic index) and desaturation activity were observed. Overall, these results indicate that dietary GCE increased the milk fat content, resulting in higher FCM and ECM with minimal effect on milk FA profile.

The benefits of feeding *Himantalia elongata* to dairy cattle on nitrogen utilisation and end-product quality
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Brown seaweeds contain phlorotannins which can positively impact nitrogen (N) utilisation in ruminants. Seaweed provision can increase iodine (I) content in milk but the effects on milk fatty acid (FA) profile have not been reported. This study aimed to evaluate the impact of the dietary inclusion of *Himantalia elongata* (HE) on N efficiency, I content while being the first study to explore the FA profile of the resulting milk. Nine late lactation multiparous Holstein-Friesian cows were evaluated in a 3 (diet) x 3 (period) Latin square design experiment with 21 days/period. The 3 groups were balanced by parity, body weight, and milk yield. Treatment diets (DM basis) included 40% concentrates and i) 60% grass silage (GS) (CON), ii) 56% GS and 4% HE extract (XHE) and iii) 56% GS and 4% HE (HE). DM intake of CON was significantly lower than XHE and HE (P<0.05) with no differences noted for milk yield (P>0.05). N intake was significantly lower in CON (P<0.05) but the XHE and HE cows had a significantly lower urine N output (g/day) (P<0.05), urinary N:N intake (g/g) (P<0.05) and manure N:N intake (g/g) (P<0.05). XHE and HE increased the combined EPA and DPA content (P<0.05). A significant positive correlation observed between I intake and blood I concentration (P<0.05). Feeding HE or XHE to dairy cows will result in I-enriched milk which can be used to increase I intake in vulnerable demographics in addition to improving N utilisation in dairy cattle.

Effect of the addition of plant galactogogues in the dairy cows rations on the quality parameters and protein profile of milk

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The study aimed to determine the impact of plant galactogogues, fenugreek and milk thistle, addition to the dairy cows rations on milk yield, its' quality and composition, including the fatty acid profile of the milk fat. The experiment involved 50 cows (70-100 days of lactation). Experimental cows were selected as analogues based on age, milk yield and stage of lactation. 5 experimental groups were formed (n=10 each): C-control, fed a standard feed ration; O1-experimental 1, supplemented with 150g of milk thistle seeds; O2-experimental 2, supplemented with 300g milk thistle seeds; K1 and K2 -experimental groups 3 and 4, receiving 150g and 300g of fenugreek seeds, respectively. The observation period lasted 6 weeks. In each week of observation, feed intake and production effects (feed utilization, milk yield) and animals health were monitored. The chemical composition and feed nutritional value were evaluated. In milk, the basic composition and total number of somatic cells and bacterial cells were determined. The fatty acid profile of milk fat was analyzed by gas chromatography. The obtained results indicate that fenugreek and milk thistle have a positive effect on lactogenesis, milk yield and milk composition. However, this effect was dose-dependent. The composition of the fatty acid profile of the milk fat of the cows of the experimental groups confirm the possibility of modification of milk fat composition by proper supplementation. The addition of fenugreek seeds and milk thistle to the ration of cows improved the health-promoting properties of milk fat.

Does feeding hay versus grass silage impact milk composition?

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Feeding dairy cows grass conserved as either hay or silage could affect the characteristics of milk, especially those important for hard cheese production. The aim of the present study was to provide an insight into the effects of feeding hay or silage derived from the same parent grass material on milk composition with regard to vitamins B1, B2, B6, B12, and E, as well as sensory properties. 18 lactating Holstein cows were divided into two feeding groups and fed either hay or grass silage, as well as 3.7 kg (DM) concentrate per cow and day. Data collection (21d) commenced after an adaptation period (14d) to the altered feeding conditions. Average milk yield was 26.6 kg/d during the data collection period. Milk samples were collected from each cow on days 2, 12, and 20 for chemical and sensory analysis. Statistical analysis of the mean vitamin contents was performed with SAS 9.4 (proc glm), including the fixed effect of the treatment (hay, grass silage) and milk yield as a covariate. Results revealed no significant difference for vitamins B1, B2, and B6. Concentrations of vitamins B12 and E were significantly increased ($P < 0.01$) in the milk from cows receiving grass silage. Colour and odour intensity were found to be higher in the milk from silage-fed cows during sensory analysis.

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Theatre 5

Cheese quality from grazing sheep supplemented with agro-industrial by-products

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This study evaluated the effect of dietary agro-industrial by-products (AIBP) on sheep cheese quality traits. During 42 days, two groups of 12 grazing Comisana ewes were supplemented with a commercial concentrate (CTRL) or a concentrate (EXP) containing a mix of AIBP (bran, distillers, rice husk, olive pomace, tomato peels) and alfalfa hay. Bulk milk from the two groups was collected to produce CTRL and EXP cheeses. Data about cheese fatty acids (FA) and antioxidant capacity were analyzed using a mixed linear model considering the fixed effect of diet and the random effect of cheese wheel. For lipid oxidation (TBARS), color, and sensory properties measured on cheese slices, the fixed effect of refrigerated storage time (0, 3, 7 days) and diet \times time were added to the model. EXP cheese showed a higher content of FA beneficial to human health and a lower content of saturated FA, resulting in a lower atherogenic index. The antioxidant capacity of EXP cheese was higher than CTRL, indicating the possible transfer of antioxidants from AIBP to cheese. This could explain why, despite the higher content of unsaturated FA in EXP cheese compared to CTRL, the cheeses did not differ for TBARS over storage time. EXP cheese showed higher yellowness and chroma. In a triangular test, EXP and CTRL cheeses were found to differ in appearance, but not in smell and taste. Also, the sensory analysis revealed no off-flavors in both cheeses. Dietary AIBP can be a viable strategy to improve cheese quality without adversely affecting sensory quality.

Dairy cows' diet has a marked impact on vitamin B content in milk but less in yogurts

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Cows' diet can have an impact on B vitamins content of milk, thus, their content may vary accordingly in yogurts. This study investigates how the contents in vitamin B1, B2, B5, B6, B9 and B12 vary between raw tank milks and the corresponding yogurts from four 10-cows herds fed different levels of grazed grass. Plain and probiotic (with *Propionibacterium freudenreichii* and *Lactobacillus Casei*) yoghurts were produced 3 times from 2 of these milks. It was observed that milk contents of vitamins B2 ($p < 0.001$) and B9 ($p < 0.10$) are higher (18% and 50%) when cows are fed grazed grass whereas it is the opposite for vitamin B5 ($p = 0.001$). Significant correlations were observed between matrices for a given vitamin but only vitamins B6 and B9 contents tended to be higher in yogurts from cows fed diet containing high percentage of grazed grass. Probiotics effect was limited to the vitamin B6 content that was increased. Diet composition induces variations of B vitamins content in milk but only marginally in yogurts. However, this remains to be verified on a larger number of production runs.

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Theatre 7

Ensiled artichoke bracts in beef cattle diet: meat quality during dry aging

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The aim of the study is to observe the effects of the inclusion of artichoke bract silage (BS) on the quality of beef meat. Forty-eight Holstein×Blue Belgian heifers were randomly assigned to 3 experimental groups: a control group (C) and 2 experimental groups receiving two different percentages of forage substitution with BS (S1; S2) for 60 days before slaughter. Feed rations were isoenergetic and isoproteic. After slaughter, the left loin was removed from each animal and subjected to dry aging for 42 days. At each experimental time (D0, D14, D21, D28, D35, D42) a 30 mm rib was cut from each loin for the colorimetric, physicochemical, and oxidative analysis. Diet affected lightness, which was higher in S1 and S2 compared to the C group at D42 (37.65 and 37.7 vs. 35.15) ($P < 0.01$). Water-holding capacity (WHC) increased from D0 to D21 in S1 ($P < 0.01$) reaching higher values compared to those of the C group ($P < 0.01$) (83.28 vs. 78.28 %, respectively). Cooking loss was higher in S1 and S2 compared to the C group at D28 and D42 (29.96 and 30.23 vs. 25.68 %) ($P < 0.01$). Oxidative patterns showed no differences based on diet ($P > 0.05$). TBARS increased from D0 to D14 only in C group ($P < 0.01$). Shear force decreased from D0 to D14 in all groups, remaining constant in the subsequent time points ($P < 0.01$). Textural parameters were not affected by the diet nor the aging time ($P > 0.05$). Based on the preliminary results, BS does not compromise the qualitative characteristics of meat; rather, it could enhance its WHC and oxidative stability. Acknowledgements: This study was carried out within the Agritech National Research Center.

The inclusion of almond hulls in lamb diets improves the fatty acid composition of intramuscular fat
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The aim of this work was to evaluate the effect of partial replacement of cereals with increasing almond hulls (AH) levels in lambs' diet on growth performance, total lipids and fatty acid (FA) composition of intramuscular fat. 24 ram lambs were assigned to one of 3 diets, in which cereals were stepwise replaced with AH, reaching 0, 9 and 18% (dry matter basis) of AH in diet. All diets included 40% dehydrated lucerne and 5% soybean oil. After 7 days of adaptation, the average daily gain (ADG) and feed intake were monitored. Total lipids were extracted from Longissimus thoracis (LT) muscle, with dichloromethane:methanol (2:1,v/v), transesterified into FA methyl esters and analysed by gas chromatography with flame ionization detection. Data was analysed using ProcMixed (SAS). The partial replacement of cereals with AH up to 18% did not affect the ADG (349 g/d), but linearly increased feed conversion ratio ($P=0.004$). Total lipid content of LT was not affected by diet. Meat contents of t11–18:1 ($P=0.049$) and c9,t11–18:2 ($P=0.004$) increased linearly with increasing levels of AH in diet. Partially replacing cereals with AH in the diet increased the health-beneficial FA in lamb meat, maintaining high growth performance. Funding: SubProMais (PDR2020-101-030988, PDR2020-101-030993); Portuguese Foundation for Science and Technology under projects UIDB/00276/2020 (CIISA), LA/P/0059/2020 (AL4AnimalS), UIDB/05183/2020 (MED), LA/P/0121/2020 (CHANGE), and PhD grant awarded to LC (2020.05712.BD)

Feeding hazelnut skin and linseed to lambs: effect on meat oxidative stability

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This study aimed at investigating the effect of the combined inclusion of hazelnut skin and extruded linseed, in the diet of lambs as partial replacer of corn and soy, on the oxidative stability of meat. Forty male lambs were randomly assigned to 4 groups and fed ad libitum for 60 days with: a conventional diet (CON); a diet with 15% of hazelnut skin (H); a diet with 8% of linseed (L); a diet with 4% of linseed and 7.5% of hazelnut skin (H+L). Fatty acid (FA) profile, fat-soluble vitamins, and hydrophilic antioxidant capacity were assessed in fresh meat, while colour and lipid oxidation were evaluated over 7 days of refrigerated storage. No effects of dietary treatment were observed on meat colour and metmyoglobin development ($P>0.05$) during storage. Similarly, antioxidant capacity assays (i.e., TEAC, FRAP, FICA, Folin-Ciocalteu, DPPH) performed in meat hydrophilic fraction showed no differences between treatments ($P>0.05$). The inclusion of H+L enriched the intramuscular fat of α -linolenic acid (3.75-fold) and other n-3 FA (+40%), notable for being health-promoting but also for their higher oxidative susceptibility. Despite a higher polyunsaturated FA content, lipid oxidation was reduced during storage in the H and H+L groups ($P=0.001$). Vitamin E content was higher ($P<0.001$) in meat from lambs fed H and H+L. The lack of the differences in the hydrophilic extract and the higher content of vitamin E could suggest that the latter compound may had a fundamental role in delaying lipid oxidation. In conclusion, feeding H+L improved the meat oxidative stability by delaying lipid oxidation probably due to vitamin E contained in hazelnut skin, even though the FA profile was improved with a higher content of polyunsaturated FA provided by linseed.

Dietary pistachio skin improves lipid profile and lipid oxidation of lamb

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This study investigated the meat quality from lambs fed with pistachio skin. Fourteen lambs were fed with a conventional concentrate (C), while thirteen animals received the same concentrate supplemented with 12% (dry matter basis) pistachio skin (P) for 60 days. The dietary treatment did not affect animal performance, carcass trait and intramuscular fat content ($P>0.05$). Polyunsaturated fatty acids (PUFA) of meat were increased (+10.83%) by pistachio diet ($P=0.047$) as well as linoleic acid (+12.43%; $P=0.032$). Although PUFA and n-6/n-3 ratio were lower in the control treatment ($P=0.029$), atherogenic index was not affected by dietary treatment ($P=0.496$). Concerning color, the Pistachio diet limited the browning of meat as KS572/KS525 was higher in P treatment (+1.57%; $P=0.009$) compared with control. Moreover, feeding P reduced lipid oxidation (TBARS values) after 7 days of storage in meat ($P=0.045$), despite the higher content of PUFA sensitive to oxidation. Probably the higher vitamin E in meat preserved meat from oxidation (+136%; $P<0.001$). In summary, the results indicate that the dietary inclusion of 12% (dry matter basis) pistachio skin has no negative impact on animal performance. Furthermore, it leads to an increase in meat concentration of polyunsaturated fatty acids and oxidative stability of lipids. This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (Piano nazionale di ripresa e resilienza (PNRR) – Missione 4 Componente 2, Investimento 2, Investimento 1.4 – D.D. 1032 17/06/2022, CN000002).

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Theatre 11

Influence of a waxy corn diet and sex on growth performance, carcass characteristics and meat quality of heavy pigs

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Although genetic selection is addressed to obtain leaner pigs, the dry-cured ham industry requires fatter carcasses. Utilizing highly digestible waxy corn could contribute to solve this issue by enhancing fat deposition. This study aimed to assess the impact of a waxy corn diet and sex on heavy pig growth, carcass traits, and meat quality. Twenty-four pigs, 12 gilts and 12 barrows, were equally divided into non-waxy (CON) and waxy (WAX) corn diet groups. Experimental diets have been formulated to be isocaloric and to provide the same amount of starch. In the range of body weight from 77.9 ± 5.2 kg to 173.1 ± 5.2 kg (slaughter weight), significant effects on average daily gain (ADG) and feed conversion ratio (F:G) were observed ($P<0.01$). The interaction diet x sex was significant being ADG of WAX barrows greater than CON diet-fed pigs or WAX gilts. The WAX diet significantly increased hot carcass weight and yield. Again, gender influenced carcass performance, with barrows showing superior performance compared to gilts. Backfat thickness and lean meat percentage were affected by diet, sex, and their interaction ($P<0.05$). WAX barrows had increased backfat thickness and reduced lean meat compared to CON barrows or WAX gilts. Meat quality traits such as color, pH24h, and water-holding capacity were not negatively affected by diet or sex. Intramuscular fat content and classes of fatty acids were unaffected by diet, but barrows had higher intramuscular lipids. Barrows had higher saturated fatty acids, while gilts had higher PUFA. The diet influenced several individual fatty acids belonging to the n-3 and n-6 series. In conclusion, the waxy corn diet effectively increased carcass adiposity in barrows without compromising meat quality.

Chemical and physical quality traits of meat from heavy pigs fed diets with a partial to total replacement of soybean meal with spirulina powder

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This study investigated the impact of replacing soybean meal (SBM) with *Spirulina platensis* (SP) on meat quality of heavy pigs. Eighty-8 gilts and barrows were fed from 52±4 to 175±6 kg body weight a control diet (C), based on cereals and SBM, or 3 experimental diets, formulated by replacing 33% (SP1), 66% (SP2) or 100% (SP3) SBM with SP (2 pens per diet). Feeds were isoenergetic, isoproteic, and iso-amino acidic. Samples of loin, lard and subcutaneous fat from hams were collected and analyzed for pH, color, shear force, proximate composition, and fatty acid profile. Data were analyzed with a linear mixed model including the fixed effects of sex, diet, and their interaction, and the random effect of pen within diet. Diet did not affect meat quality traits, with few exceptions: CIE a*, CIE b* and Chroma were greater in SP3 than in C (P<0.05); likewise was CIE b* in the ham's subcutaneous fat from SP3 than from C (P<0.05). Similarly, loin cooking losses was 7.8 % greater in SP3 than in C (P<0.05). A diet-related effect was also found for arachidonic acid in ham's subcutaneous fat (P<0.05), with greater content for SP2 and SP3 than for C. In conclusion, confirming the evidence found for growth and carcass traits, the full replacement of SBM with SP exerted minimal effects also on meat quality traits. Further research is needed to assess the impact of SP on the technological and sensorial quality of the dry-cured ham. This study was carried out within the Agritech National Research Center and received funding from the European Union Next-Generation EU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022).

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Theatre 13

Dietary leucine modulate pork quality via adjusting muscle fiber type conversion and intramuscular fat deposition in Huanjiang mini-pigs

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This study was designed to investigate the effects of leucine (Leu)-supplemented diets on meat quality, muscle fiber types, and intramuscular fat (IMF) content modulated by gut microbiota composition in Huanjiang mini-pigs, a Chinese local rare pig breed with delicious meat quality. The total amount of 192 male Huanjiang mini-pigs with average body weight of 9.80 ± 0.05 kg (mean ± SEM) were randomly allotted to one of four dietary treatments with 8 replicate pens per treatment and 6 pigs per pen: a control group (a basal diet), and three treatment groups supplemented with different concentrations of Leu (0.5%, 1%, 1.5%). The experiment had duration of 60 days until the average final body weight of Huanjiang mini-pigs was 30.38 ± 0.21 kg. All data were analyzed by one-way analysis of variance (ANOVA) using the IBM SPSS 26 software, followed by Tukey's multiple-range test to determine treatment effects. The results were regarded as statistically significant at P < 0.05. The results showed that compared to the control diet, the group supplemented with 2% Leu not only had higher average daily gain and carcass weight (P < 0.05), but also improved meat tenderness and juiciness by decreasing the shear force and increasing the water holding capacity (P < 0.05). Particularly, this treatment showed higher alpha diversity of gut microbiota (P < 0.05), as well as higher mRNA expression levels of acetyl-CoA carboxylase (ACC), peroxisome proliferation-activated receptor-γ (PPARγ) (P < 0.05), increasing the composition of IMF (P < 0.05) and MyHC I (P < 0.05) in longissimus dorsi muscle (LDM). Our findings suggest that specific concentration of Leu could improve meat quality of Chinese local pigs, by modulating IMF deposition and fiber type switching, and provide a molecular basis for cross-regulation to reveal the nutrient regulatory network of pork quality.

Milk yield and composition of Cinisara grazing cows supplemented with a low- and a high-polyphenols faba bean varieties

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The crude protein (CP) of fresh grass is generally characterized by a high degradability which can lead to reduced efficiency of its utilization. The presence of a moderate level of polyphenolic substances in the diet of grazing animals could help to overcome this problem. The aim of the study was to evaluate the effects of grazing supplementation with 2 varieties of faba bean with different polyphenols content on milk yield and quality in Cinisara grazing cows. Fifteen cows, homogeneous per days in milk (61 ± 29) and milk yield ($12.9 \text{ kg} \pm 3.2$) fed for 20 h/d on a mixed pasture (CP 17.9% DM) and supplied with grains (1.5 kg of barley, 1.5 kg of oats, 1.5 of wheat) and a mixed hay (CP 10.2% DM) were distributed in two experimental groups and supplemented as follows: low polyphenols group (LP, 8 cows), 2 kg/d of faba bean var. Torrelama (4.4 mg GAE/g DM; CP 28.4% DM); high polyphenols group (HP, 7 cows), 2 kg/d of faba bean var. Fanfare (16.4 mg GAE/g DM; CP 28.9% DM). The trial lasted 4 weeks during which individual milk yield was measured at the end of each week and individual milk samples from the two daily milkings were collected. Milk samples were analyzed for gross composition and fatty acid profile. Data were analyzed by ANOVA for repeated measures. The HP supplement caused an increase in milk yield and casein level, compared to LP supplement (respectively 16.2 vs 14.5 kg/d, $P=0.051$; 2.71 vs 2.61% , $P=0.034$). No diet effect was evident on milk fatty acid profile. The results seem to suggest an improvement in the efficiency of pasture utilization linked to dietary supplementation with the variety of faba bean richer in polyphenols.

Study of the effect of Fabaceae plants on the meat quality of heavy fattening pigs Złotnicka Spotted breed
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The purpose of the study was to determine for slow-growing Złotnicka Spotted pigs the appropriate composition of feed mixtures containing Fabaceae plants, ensuring the most favourable fattening efficiency for this breed and high meat quality, confirmed by dietary and technological parameters. The experiment was carried out on 96 fattening pigs of the Złotnicka Spotted breed. Animals were divided into 4 feeding groups: group A – control cereal-soybean mixture, and 3 experimental mixtures containing different types of domestic protein sources (groups B – lupin, C – faba bean and D – peas). During fattening, animals between 30 and 80 kg body weight were fed a grower diet, and from 80 kg body weight to the established final slaughter weight (140 kg) a finisher diet. All diets were isoproteic and isoenergetic within the fattening phases. After the animals were slaughtered, the parameters of selected carcass quality traits were determined, the meat content of the carcass was measured with an optical probe device, and meat samples (longissimus lumborum) were taken for chemical analysis and evaluation of meat quality parameters. There was no effect of nutrition on fattening and slaughter traits. It was found that heavy pigs fattened with experimental mixture C and mixture D were characterized by meat with the most favourable values in terms of PUFA n-6/n-3 fatty acid ratios. Heavy pigs fattened with mixture C were also characterized by the most favourable meat oxidative stability (TBARS) among all feeding groups. The work financed from the National Research Institute of Animal Production (Task no. 01-11-04-11).

Effects of dietary supplementation with enriched olive cake on meat quality traits of Limousine bulls

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Olive cake (OC) is considered an agro-industrial waste, but the high content of valuable metabolites makes it a promising feed integration. Therefore, this study aimed to evaluate the effects of two levels of OC integration in the concentrate on meat chemical composition and fatty acids profile of Limousine bulls. Thirty bulls (10 × group) were blocked by body weight (BW) and then randomly assigned to 1 of 3 dietary treatments for 140 d: concentrate without inclusion of OC (CTR), concentrate with 10% (OC10), and concentrate with 15% (OC15). Individual samples of Longissimus dorsi muscle were collected 24 h after slaughter. Lipid, protein and fatty acids profile were determined. Data were analyzed with GLIMMIX procedure of SAS. No differences were detected for lipid and protein contents. Regarding fatty acids, C14:0, C14:1, C15:0, C16:0, C16:1, C17:0, and C17:1 linearly decreased with increasing levels of OC supplementation, where meat of (OC15) group had lower levels than (OC10) group and both than CTR (P<0.05). Whereas, C18:0, C18:1n9, C18:2n6, C18:3n3, C20:0, C20:4n6, C20:5n3, and C22:5n3 linearly increased by increasing levels of OC integration, with OC15 samples having greater levels than OC10 and both than CTR (P<0.05). In addition, long-chain fatty acid C20:3n6 linearly decreased (P<0.05). Fatty acid classes were linearly affected; in fact, SFA decreased, whereas MUFA, PUFA, n3, and n6 increased by increasing levels of OC supplementation (P<0.01). Regarding nutritional indices, atherogenic and thrombogenic ones linearly decreased (P<0.01). Concluding, OC can be considered a functional component in bull diets with positive influence on UFAs, providing healthy animal products and at the same time being a strategy for the sustainability of the animal food chain included in the concept of circular economy.

Session 62

Poster 17

Effect of incorporating protected soya lecithin in a high oleic finishing diet of oxen on fatty acid profile of subcutaneous and intramuscular fat

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In ruminants' diets enriched with fat, lecithins are used to facilitate the emulsification, improving absorption in the digestive tract, therefore increasing production efficiency and modifying fatty acid composition. The aim of the study was to evaluate the incorporation of protected soya lecithin in oxen fed with high oleic diets, on meat composition and fatty acid profile of the subcutaneous and intramuscular fat. Eight oxen 4-6 years old were used to test two experimental diets during a finishing of 12 months (four animals each group). CON (control diet) included oleic oil at 2,8% and SOY (control with 0,5% soya lecithin). After slaughter, subcutaneous fat and L. lumborum muscle was removed. Chemical composition of muscle and fatty acid profile both in subcutaneous and Intramuscular fat was analysed. Data was subjected to on-way analyses of variance (diet). SOY presented lower protein values than CON (20.7 vs 19.6%) and intramuscular fat content 30% higher than CON (17.3 vs 12.2%). Regarding fatty acid profile of subcutaneous fat, SOY showed high MUFA percentage (66.1 vs. 71.3) and lower SFA (31.4 vs. 27.75). However, the inclusion of lecithin, did not resulted in differences in intramuscular fatty acid profile, probably due to that animals received diets rich MUFA, desaturation is already very high and the effect of lecithin may be masked. As a conclusion, the incorporation of soy lecithin in oxen diet resulted in a higher fat content, and in higher MUFA content in subcutaneous fat, so it is seemed to be a tool to improve production efficiency and modifying fatty acid composition. This work has been funded by Funtional Nutrition S.L

Impact of protein source on pork quality and production parameters: a comparative study of biorefined grass-clover protein and soy-cake protein for organic growing-finishing pigs

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Aim: This study assessed the impact of feeding organic pigs biorefined grass-clover protein or soy cake protein from 30-155 kg. The two concentrates were formulated as isoenergetic and provided similar digestible lysine levels. The study involved 270 pigs, and production parameters, such as daily gain, feed consumption, feed efficiency, blood parameters; meat percentage, meat quality, and eating quality, were measured. **Design:** The experiment took place at the Organic Platform at AU-Viborg, Denmark, with DLY or LYD pigs of DanBred-origin weaned at 7 weeks and introduced into the experiment at 30 kg. The rearing period lasted 13-14 weeks. Each pig received 200 Scandinavian Feed Units (FU) of concentrate and approximately 20 FU of roughage. **Key results** include Biorefined grass-clover protein in the concentrate increased the meat percentage compared with soy-cake protein. Soy protein can be replaced with grass protein without negatively affecting daily gain or feed consumption. Soy-based concentrate leads to a higher cooking loss and showed a tendency for more structures on the cutting surface in pork than biorefined grass protein. Grass protein led to higher nitrogen deposition than soy protein, suggesting a more suitable amino acid composition. No differences were observed in meat pH, intramuscular fat amount, texture after 6 months of storage, acidity, harshness, meat flavour, metallic taste, bitterness, juiciness, chew resistance, chewing time or pork tenderness between protein sources. Higher content of omega-3 fatty acid alpha-linolenic acid and lower vitamin E in meat from biorefined grass-clover protein-fed pigs

Seasonal quality variation of Bresaola from grass-fed Angus beef

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Bresaola is a cold cut obtained from bovine meat. The seasonal variability of the diet in a grass-fed system may affect meat composition, which in turn may result in a variation in meat product quality over the year. Therefore, this study assessed the effect of the feeding season on the quality traits of bresaola from grass-fed Angus beef. To this aim, meat from 29 animals from the same organic farm was used to produce individual bresaolas. The bresaolas were divided in 3 groups according to the main diet in the 2 months before slaughter: Fresh pasture (FP= 12), dry pasture (DP= 7), and hay (HA= 10). Data on fatty acids (FA), myoglobin, and antioxidant capacity (TEAC) were analyzed using a mixed model, with the random effect of the animal and the fixed effect of the diet. For lipid oxidation (TBARS) and color analysis (CIELab space), the fixed effect of time (0, 3, and 7 days of refrigerated storage) and time × diet were added to the model. Diet did not affect FA and total myoglobin, whereas metmyoglobin was higher for DP than FP and HA. FP bresaola showed the highest TEAC. Color indices deteriorated over storage time, with a* worsening at day 3 and L after 7 days. Yellowness (b*) and hue angle (h*) were affected by time × feeding period. After 3 days of storage b* decreased for DP and HA, while remaining stable for FP. The h* index, was stable during storage for FP and HA, while it increased at day 3 of refrigerated storage and decreased at day 7 for DP. TBARS increased over storage time despite groups, but the feeding season was ineffective. Seasonal variation was observed in bresaola quality from grass-fed animals, with DP showing worse values than HA and FP.

Effects of dietary supplementation of oregano essential oil to feedlot cattle on their meat quality and muscle fiber histomorphometry

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Essential oils are common ingredients of feed supplements in animal nutrition. The notion is that they contribute positively in quality characteristics of meat and enhance its shelf life. The objective here was to assess the role of Oregano Essential Oil (OEO) as a feed additive on quality characteristics of meat produced by feedlot calves. Sixty-eight Limousine bulls were allocated into two groups (n= 34), designated as control (C) and test (T). Group C was fed a typical commercial diet whereas T was fed the same diet supplemented with OEO at 50mg/kg DM, for 90 days. After this period, all bulls were slaughtered and their carcasses were assessed for microbial contamination. Steaks (13th rib), from all carcasses were obtained to assess meat color, texture, pH, chemical composition and oxidation stability as well as fatty acids and amino acids profile. Fiber histomorphometry (Minimum Feret Diameter) was assessed in formalin-fixed muscle samples. Moreover, sensory evaluation of meat samples was done by a panel of trained consumers. Data were analyzed using SPSS software; significance level was set at P<0.05. Microbial counts of examined carcasses were generally below the limits set by EU Commission Regulation No. 2073/2005. Meat physicochemical properties, organoleptic characteristics, texture and color did not differ between the groups (P>0.05). However, meat oxidation stability was higher in carcasses of T group (P<0.05). The Minimum Feret Diameter was similar between the two groups (P>0.05). The results suggest that dietary supplementation of OEO to feedlot cattle has positively impact the quality of produced meat. This research has been co-financed by the European Regional Development Fund of the European Union and Greek National Funds through the Operational Program Central Macedonia 2014-2020 (KMP6-0280294; BellasQualityMeat).

Effects of the incorporation of sainfoin as hay or included in the concentrate on carcass and meat quality of young bulls

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The inclusion of sainfoin (*Onobrychis viciifolia*) in ruminant diets has been promoted because of its high quality and its agronomic and environmental benefits. To optimise its use, it must be preserved as hay or pellets, but its inclusion in the diet of intensively fed fattening cattle could affect carcass parameters and meat quality. Forty Montbeliard young bulls (age: 117 d, body weight (BW): 188 kg) were ad libitum fed four diets for 28 weeks: concentrate + straw (C+straw), concentrate + sainfoin hay (C+SFhay), concentrate including 15% sainfoin + straw (CSF+straw) and concentrate with 15% sainfoin + sainfoin hay (CSF+sfhay). The effect of the diet on slaughter BW, hot carcass weight (HCC), dressing percentage (DP), conformation (1-18), fatness score (1-5), subcutaneous fat colour, and meat pH, chemical composition and colour was studied. The diet affected slaughter BW (498, 490, 466, 461 kg, for C+straw, CSF+SFhay, C+SFhay and CSF+straw respectively, P<0.01), but had no effect on HCC (274 kg), fatness score (2) and fat colour (lightness (L*): 73.3; redness (a*): 4.0, yellowness (b*): 11) (P>0.05). The diet affected DP (56%, 57%, 59% and 59%, respectively; P<0.001); and conformation (6.0, 6.0, 5.8, 6.5, respectively; P<0.05). The diet did not affect meat pH (5.6) and protein content (22.6% fresh matter (FM), P>0.05), but tended to affect fat (3.4, 3.3, 2.7 and 3.0% FM, respectively; P<0.10). Meat L* was affected by the diet (39.3, 41.0, 39.5, 39.3, respectively; P<0.05) but a* (13.1) and b* (8.1) were similar (P>0.05). Thus, despite the differences in slaughter BW, sainfoin inclusion mildly affected most carcass and meat quality parameters

Antibacterial activity and functional characteristics of lactic acid bacteria isolated from green tea and their application in producing dry-cured ham

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This study aimed to investigate the antibacterial activity of probiotic microorganisms that are usually used to produce safe dry-cured hams and their application in ham production. Thirty strains of lactic acid bacteria (LAB) were isolated from 44 food sources, including green tea, strawberries, and Korean beef. The obtained isolates and their cell-free supernatants were tested for their antibacterial activity by agar well diffusion assay. Among these, nine isolates exhibited antibacterial activity against five food-borne bacterial pathogens (*Listeria monocytogenes*, *Salmonella enterica*, *Escherichia coli*, *Bacillus cereus*, and *Staphylococcus aureus*), as well as against two toxin-producing fungal pathogens (*Aspergillus niger* and *Fusarium solani*). These nine strains were further evaluated for functional characteristics, including salt tolerance, low temperature tolerance, drying tolerance, and nitrite tolerance. *Lactiplantibacillus plantarum* G2 and *Levilactobacillus brevis* H8 demonstrated superior tolerance to salt, nitrate, and low temperatures during the simulated stages of fermentation and ripening of dry-cured ham. Subsequently, these *Lactobacillus* strains were applied to Lonzino, and a 90% reduction in the total number of aerobic bacteria and fungi in the G2-treated group was observed, compared to the untreated group, at the 7th month, which marks the completion stage of fermentation. In conclusion, these strains showed antimicrobial activity against harmful microorganisms found in dry-cured ham, which could contribute to improving the hygienic quality of the product and preventing possible diseases.

Effect of feed mixtures contamination with low doses of antibiotics on meat quality of broiler chickens

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The aim of the study was to determine the effect of low doses of antibiotics in the diets with that mimic the cross-contamination of non-medicated feed, given to chickens during the whole rearing period on meat quality. One-day old broiler chickens were randomly divided into seven groups and fed ad libitum with the basal complete starter (1–21 d) and grower/finisher (22–35 d) diets containing colistin (group I), doxycycline (II), flumequine (III), thiamphenicol (IV), tiamulin (V) or tilmicosin (VI) at the doses of 1% of the recommended concentration in the medicated feed, while the control group (VII) received antibiotic-free diets. On day 35 of the experiment, breast and thigh muscles were collected to determine pH, conductivity, Lab colour, drip loss, cooking loss, shear force, texture parameters (hardness, springiness, cohesiveness, gumminess, chewiness), chemical composition (dry matter, crude protein, crude fat, crude ash), cholesterol content, fatty acid (FA) profile and fat oxidative stability (TBARS, on the slaughter day and after 3 months of storage at -18oC). The results were analysed by one-way analysis of variance and Duncan's test. The breast muscles did not differ in terms of physicochemical parameters, while the leg muscles differed depending on the tested antibiotic in the diet in terms of cooking losses and texture, with the most favorable results in the control group, and the least in g III. There was no significant effect of low-dose antibiotic feed contamination on the TBARS value and chemical composition of the muscles, when compared to control group. However, some differences in the FA profile were indicated. In conclusion, the antibiotic contamination did not affect the meat quality negatively. This research was funded in whole by the National Science Centre, Poland, under project no 2021/41/B/NZ9/04114.

The use of grape pomace in rabbit diet improves the oxidative stability of the meat
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The oxidation of lipids is one of the main factors that limit the quality and the acceptance of meat and meat products. In recent years, attempts have been made to enhance the stability of meat through dietary strategies to reduce the use of artificial additives in foods. The objective of this study was to evaluate the effect of the inclusion of grape pomace (GP), a byproduct rich in antioxidant compounds, in the diet of rabbits on fatty acids (FA) profile and oxidative stability of the longissimus dorsi (LD). Forty eight rabbits at 55±3 days of age (mean±SD) were used in the trial. The animals were divided into three groups: control group (CRT), feeding a basal diet and treated groups fed the basal diet supplemented with 5 g/day (GP5) and 10 g/day (GP10) per head of GP. The trial lasted 3 weeks and at the rabbits were slaughter at 76 days of age. The LD muscle was dissected and analysed for pH, chemical composition, FA profile and Thiobarbituric acid reactive substances (TBARS). Data were analysed by one way ANOVA with the diet as fixed effect. The live weight and the ADG were not influenced by the diet ($P > 0.05$). The inclusion of GP in the diet of rabbit did not influence the proximate chemical composition. The most abundant fatty acid in rabbit meat was the palmitic acid (C16:0), followed by linoleic acid (C18:2n6) and oleic acid (C18:1c9) that represent together the 76% of the total FA. The GP supplementation did not affect the FA profile but led to a linear decrease of the TBARS values expressed as mg of MDA per kg of dry meat. The values of mg of MDA per kg of dry meat were 0.77, 0.62 and 0.33 for CRT, GP5 and GP10 groups, respectively. In conclusion, 5 and 10 g/d of grape pomace can be used in rabbit diet during the fattening period without negative effect on growth performance and meat quality. Also, the inclusion of this by product in the diet of these animals could be a good nutritional strategy to improve the oxidative stability of the meat, as demonstrated by the reduction of mg of MDA. Acknowledgements: This work was financially supported by NextGenerationEU; PNRR M4C2, CN00000022 AGRITECH.

Tomato and pepper waste in sustainable laying hens diet for improving nutritional and organoleptic characteristics of eggs

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Fresh tomatoes (T) and peppers (P), waste unsold from local markets, were supplemented to laying hens diet, with the aim to evaluate their nutraceutical effect on hens performance and eggs quality. Forty ISA Brown laying hens (70-weeks-old) reared in outdoor system, were divided into 2 groups of 20 hens each and fed a basal diet (C group) or a basal diet supplemented with 300 g/kg of T + 230 g/kg of P (TP group). After 3 months, hens body weight (BW) was recorded and 15 eggs sampled from each group. Eggs weight (EW) was registered and egg mass (EM) calculated. Chemical composition and cholesterol content were determined. The color and odor profile of yolk was evaluated using an E-Eye and an E-Nose. Performance and chemical data were submitted to ANOVA and sensorial data to a DFA analysis. Results showed the positive effect of use of T and P on palatability of the diet and confirmed the nutraceutical effects of T and P, increasing BW (2,071g TP vs 1,913g C), EW (51 g/hen/day TP vs 47 g/hen/day C), and reducing cholesterol content (1,71 mg/100g TP vs 2,08 C mg/100g). The addition of T and P in the diet strongly influenced the organoleptic profile of egg's yolk. For the color profile, the E-Eye showed a clear separation of the C group from the TP group. C group was characterized by RGB color codes corresponding to shades of yellow (2416, 2400, 2144, 2128, 1872), in contrast to the TP group, which was characterized by color codes corresponding to shades of orange (1328, 1584, 1600, 1824, 1840, 1856). Even for the odor profile, E-nose showed a clear separation between the groups. The P- and T-type sensors, corresponding to hydrocarbons, head toward the TP group, probably due to the high carotenoids content of T and P. The results obtained demonstrate that principles of circular economy, can be considered useful for the production of sustainable eggs with improved nutritional and organoleptic characteristics. Research funded by the European Union – NextGenerationEU. PNRR – Missione 4, Componente 2, Investimento 1.1 – PRIN 2022 PNRR – D.D. n. 1409 del 14-09-2022. CUP J53D23018560001 – P2022SN3M9

Fattening and slaughter characteristics and meat quality parameters of merino lambs fed with fodder with herbs

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The use of natural dietary supplements based on plant substances in the nutrition of farm animals has become increasingly popular. Phytogetic feed additives are used in animal nutrition to improve their productivity by changing the properties of feed and improving the quality of animal products. The aim of the research was to investigate the effect of herbal additives added to concentrate feed on the slaughter characteristics and meat quality of Merino lambs. For the experiment, two rams group of the native Old Type Merino: C (control – 12 rams) and H (experimental – 12 rams), were fed with quality hay and given a concentrated mixture ad libitum. Group H received a herbal mixture (dried) as an addition to the concentrate feed (20g/kg of feed), containing four components each (fennel, purple coneflower (Echinacea), garlic, thyme). Vital measurements were performed (body weight, ultrasound measurements of the dorsal surface area, control of feed intake). Post-mortem evaluation of the carcass included estimation of slaughter yield, measurement of cuttings, determination of the weight of cuttings and detailed dissection. Meat samples (M. quadriceps femoris) collected after slaughter were analyzed to determine the chemical composition. There was no effect of the given feed on fattening and slaughter characteristics. The diet had an impact on the chemical composition and fatty acid content of Merino lamb meat: statistically significant differences in fat content were found. The highest fat content was found in samples from lambs from group C, while the lowest fat level was recorded in group H, which received a higher level of the herbal mixture. Statistically significant differences were noted in relation to the SFA and UFA content. The level of saturated fatty acids was lower in group H, the level of unsaturated fatty acids was the highest in group H. The PUFA-3 content was almost twice higher in the experimental group H than in group C, which did not receive the addition of the herbal mixture. It can be said that the addition of herbs to the feed had an impact on selected meat quality parameters, but did not affect the fattening and slaughter characteristics of Polish Merino lambs.

Session 63

Theatre 1

Feeding behaviour: The missing link between pig welfare and pig nutrition research

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Domesticated pigs in a natural environment spend more than 50% of their active time with foraging, exploring and feeding. The expression of this behavioural complex is very species-specific and pigs are highly motivated to display it. The inability to do so may result in damaging behaviour such as tail biting. While feeding behaviour is central for the pigs' welfare, it has been widely neglected both in animal welfare as well as in animal nutrition research. These two disciplines differ in the used methodology, their understanding of welfare and also the part of the behavioural complex of feeding they are dealing with. In animal welfare research the appetitive part is in the focus. The appetitive part includes foraging, rooting and exploring. Animal welfare researchers investigate enrichment materials such as straw or artificial tools that are supposed to satisfy the behavioural needs of pigs in this respect. The actual interaction with the provided feed itself and the suitability of the feed to satisfy behavioural needs is usually not targeted in this field. Studies originating from the animal nutrition discipline, on the other hand, deal with the very last part of the behavioural complex, the consummatory part. The focus here is on nutrient and energy intake, and thus performance, efficiency and health. The approach is based on a concept of animals that prioritizes physiological and anatomical functions over emotional or behavioural aspects. The missing overlap between these two disciplines has had consequences for the developments that have been achieved in these fields. In the presentation, we will demonstrate evidence and consequences of the missing overlap as well as potential solutions.

When do ad libitum fed lactating sows choose to eat?

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To better understand the intrinsic feeding behavior of lactating sows, 51 animals across two Austrian farms were fed via a sow controlled ad libitum feeding system (ADLIB) and their feeding patterns monitored from lactation day six until weaning. Sows pushed a metal rod to trigger the delivery and digital recording of small quantities of feed (50 g). The ADLIB sows were housed in the same farrowing rooms as limit-fed sows on a fixed feeding schedule of 6:00, 12:00 and 16:00. Average feed disappearance for ADLIB sows was 6.4 kg/day and was consumed across the whole day; although with peaks at 2:00, 6:00, 12:00 and 16:00 suggesting some entrainment of ADLIB sows to the fixed feeding schedule. Peaks in the feeding patterns of older sows were more well defined compared to gilts and could result from sows being familiar with fixed feeding regimes during previous lactations. In summer, feed disappearance was less synchronized and shifted to the night as the 12:00 feeding peak disappeared. Taken together, these findings demonstrate an intrinsic feeding pattern more similar to their natural foraging behaviors that contrasts the experience of sows in fixed feeding regimes. Lactating sows eat small portions of feed over the whole day and will adapt their feed intake based on extrinsic factors such as environmental conditions like ambient temperature and social facilitation where animals are motivated to eat together. This work highlights new opportunities to better match lactating sow feed intake to her needs.

Session 63

Theatre 3

Piglet suckling behaviour in relation to fibre type in the maternal diet

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The present study aimed to determine whether the dietary fibre type in the maternal diet and the suckling position influence piglet growth between birth and 70 days (d) of age. From d85 of gestation to weaning, 40 sows were fed a gestation and lactation diet containing either 4% of oat hulls (OH) or 4% of inulin (IN). Piglet body weights were recorded on different days until d70 of age. On d3 and d17, the piglets were observed during two suckling sessions and were assigned to a suckling position on the sow's udder: A (anterior), M (middle), P (posterior), D (different position between the two suckles). Suckling behaviour was recorded on d10. Suckling area fidelity between d3 and d17 averaged 82% (± 3) and was not affected by the maternal diet. There was no interaction between suckling position and maternal diet on piglet growth at either d3 or d17. Both the maternal diet and the suckling position on d3 tended to or influenced growth from birth to d5 ($P < 0.10$). Similarly, at d17, piglets suckling on the A area had better ($P < 0.05$) growth from birth to d17 than those suckling on the M and P areas. Regardless of the suckling position, piglets from OH sows tended to grow faster ($P < 0.10$) from birth to d17 than those from IN sows. This suckling position effect persisted until weaning, but was no longer significant at d70. In contrast, the maternal diet affected growth until d70 ($P < 0.05$). At d10, piglets from OH sows tended to have fewer fights at the udder than those from IN sows. Besides a similar suckling duration, piglets from OH sows tended to suckle less often and had a longer suckling interval than those from IN sows. The present results confirm that the A area is associated with a better growth. It also showed that the fibre type in the maternal diet affects piglet growth but also suckling behaviour.

Can stocking density and feeding space influence weaned piglet performance and behaviour?

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Stocking density (SD) and feeding space (FS) is suggested to influence pig performance and behaviour. Though, the effect of these on piglets is unknown. Two trials (T) were performed: T1 consisted of low SD (LSD; 0.57 m²/piglet) and moderate SD (MSD; 0.38 m²/piglet) with 24 replicates (pen; boar: gilts in 1:1) each, and T2 consisted of 3 FS (3FS) and 6 FS (6FS) both at SD of 0.57 m²/piglet with 12 replicates each. Piglets were not tail-docked and were fed a commercial diet which was available ad libitum. Piglets in T1 were weaned (day (d) 0) at d29.8 (SD = 0.55) and 8.38 kg (SD = 0.53); piglets in T2 were weaned at d31.4 (SD = 1.66) and 7.31 kg (SD = 0.377). Piglets were individually weighed at d0, d14, and d35 PW (end). Feed intake was recorded between d0-14 and d14-35 PW. Faecal consistency (FC) was recorded two times a week and tail damage was scored at d35. The LSD piglets grew faster between d0-14 (P=0.04), d14-35 (P=0.02), and between d0-35 PW (P=0.05); ate more between d14-35 (P=0.01) and d0-35 PW (P=0.01); and had a lower FCR between d0-14 PW (P=0.02) than the MSD piglets. At d35 LSD piglets tended to be 900 g heavier than MSD piglets (P=0.06). The LSD piglets tended to have a higher FC between d0-14 PW (P=0.09) than the MSD piglets. Tail damage was not influenced by SD (P=0.12). 6FS piglets had a higher FC between d0-14 (P=0.01) and d0-35 (P=0.02) than 3FS piglets; 6FS piglets tended to have a lower FCR between d0-14 (P=0.06) than 3FS piglets. FS influenced tail damage (P=0.03) with 3FS having more intact tails (i.e. 93.8% intact tails, 6.2% moderate damaged tails, and 0.0% incomplete tails) than 6FS (i.e. 87.5% intact tails, 10.4% moderate damaged tails, and 2.1% incomplete tails). The results suggests that FS as well as SD can influence performance and FC, but also damaging behaviour.

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Theatre 5

Feed taste variety within days in nursery pigs: effect on animal's preferences and performance

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Thirty-two weaned pigs (21 days old) were used to explore the effect of taste variety within days on their productive parameters. At 28 days-old, half of the pens (n8) were exposed for 6 weeks to a commercial feed with the inclusion of 300mM of monosodium glutamate (MSG-feed; Monotony Group; MON) distributed in three feeding plates. The other half of the pens (n8; Variety Group; VAR) were exposed simultaneously to MSG-feed, the commercial feed plus 8% sucrose (SUC-feed) and the commercial feed without tastes included (CON-feed) in three feeding plates. Pig's body weight, average daily gain (ADG), average daily feed intake (ADFI), feed-to-gain ratio (FGR) and feed intake per plate in the VAR group (ADFI-P) were weekly estimated. Data were analysed with an ANOVA procedure by using the statistical software SAS®. An overall effect of feed taste was observed in ADFI-P, where the animals consumed more MSG-feed than SUC-feed and CON-feed (P<0.001) and more SUC-feed than CON-feed (P<0.001). Particularly, MSG-feed intake was higher in weeks 2, 4 and 7 (P<0.05). No differences were found in pig's performance related to experimental Groups (P>0.05), despite a higher ADG for VAR group during week 4 (P=0.05) and numerical differences observed in favour of the VAR group in most weeks measured. The strong preference for MSG could cause taste monotony throughout the days, needing new protocols with feeds of similar palatability to enhance the positive effect of sensory variety.

Peat pellets as enrichment material had positive effects on growth and intestinal microbiota of weaned piglets

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Chewable and edible enrichment material is known to increase the exploratory and feeding behavior of piglets and to improve their wellbeing. Peat is a source of fiber and humic acids, both considered beneficial for intestinal functions. The present study evaluated peat pellets (types A and B, produced by Neova Oy, Finland) as enrichment material to weaned piglets. Piglets with a mean weight of 10.2 kg were randomly divided into 30 pens, 19±2 piglets/pen, and fed with commercial piglet diets (Hankkija Oy, Finland) for 28 days. The treatments (10 pens/tr.) included Tr. 1) control (no enrichment materials), Tr. 2) peat A pellets, and Tr. 3) peat B pellets. The pellets were served ad libitum from hopper pans. On days 1 and 28, the piglets were individually weighed and sampled for faeces. The faecal samples were analysed for microbiota composition and volatile fatty acids (VFAs). Mortality and diarrhoea frequency were recorded. Data was analysed by ANOVA and Tukey's test. The mean total feed intake was 15.7 kg, 15.4 kg, and 15.5 kg/piglet for treatments 1-3, respectively. The mean total intake of peat pellets was 580 g/piglet for peat A and 480 g/piglet for peat B. Daily weight gain per piglet was 303 g, 370 g, and 353 g for the treatments 1-3, respectively ($p<0.05$), with 21% higher daily weight gain for Peat A in comparison to control. Piglet mortality or diarrhoea frequency were not affected by the treatments. In faecal samples, peat increased the concentration of VFA, especially butyric acid, increased the numbers of bifidobacteria, and reduced the numbers of *E. coli* and *Bacteroides* spp. In conclusion, peat pellets as enrichment material may be a useful strategy for improving intestinal fermentation and growth of weaned piglets.

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Theatre 7

Housing systems and dietary interventions can affect health, performance and microbiome of pigs from weaning to the growing

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The study aimed to evaluate the effect of two housing systems and a mixture of feed additives from weaning to growing phase on the health, performance and microbiome of pigs. At weaning, 150 pigs, were arranged into 6 groups into either a farm with high (H; 3 groups) or low (L; 3 groups) sanitary condition. Pigs in H and L farms received either the control diet (CO) or a diet with the inclusion of 5000 mg/kg of benzoic acid, 35 mg/kg of *E. faecium* and 500 mg/kg of muramidase until day (d) 89 (30 kg of body weight BW) (TRT-CO) or until d98 (45 kg; only benzoic acid) (TRT-TRT). Faeces and blood were collected at d42, d80 (weaning unit, T2 and T3) and d98 (fattening unit, T4). Shotgun microbiome analysis, blood formula, and oxidative stress markers were examined. At T2, but not at T3, the L farm had higher leukocytes ($P<0.01$), neutrophils ($P=0.002$), lymphocytes ($P=0.03$) and monocytes ($P<0.01$). At T2, reactive oxygen metabolites and biological antioxidant potential (BAP) were higher in the TRT_L than in the TRT_H ($P=0.05$). At T4, the L farm had higher neutrophils and lower lymphocytes ($P<0.01$) and CO_L had a higher level of BAP than all groups in the H farm, while the TRT-TRT diet had higher BAP concentration than CO and TRT-CO groups in the H farm ($P<0.05$). The TRT-TRT had a better ADG from T3 to T4 ($P<0.01$) than the CO, but only in the H farm. ADG from T1 to T4 was higher in the H than in L farm. Overall, this study confirms that both housing environment and diet modulate aspects of pig health and performance.

Dietary GABA supplementation in nursery pigs can improve object recognition memory regardless of their coping style

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Thirty-one castrated-male pigs (58 d-old) with a different coping style (HR, high resistance or LR, low resistance) were used to explore the effect of dietary γ -Aminobutyric acid (GABA) supplementation on the pig's memory through novel object recognition test. Pigs belonged to two dietary treatments, administered for 4 weeks: basal diet (CTR; n=7 HR and 8 LR) and basal diet + 300 mg/kg of GABA (GABA; n=8 HR and 8 LR). The test was conducted in the home pens, which were previously divided into two areas by a black panel. In the test area, each pig was exposed to two identical sample objects (SO) attached to the slatted floor for 10 min. After 10 min with its pen-mates, pig was returned to the test area and exposed to the SO and a completely novel object (NO) for 10 min, and its behaviour was evaluated. The object and position (left or right) of the novel object was counterbalanced. Data were analysed with a Kaplan–Meier survival method and ANOVA procedure by using the statistical software Rstudio. No interactions were found between coping style and treatment across variables. GABA group had a lower latency to contact NO, spent more time touching the NO and less time touching the SO than the CTR group ($P<0.05$). In addition, GABA group had a higher discrimination index ($[(\text{Time NO}-\text{Time SO})/(\text{Time NO}+\text{Time SO})]$) than the CTR group ($P=0.002$) and tended to exhibit more play behavior ($P=0.07$). Regarding the coping style, HR pigs spent more time touching the NO ($P=0.035$) than the LR pigs. In conclusion, coping styles do not differ in discriminating a new object from a familiar one. However, GABA-supplemented pigs showed a preference for exploring the novel object, suggesting a potential impact on cognitive function.

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Theatre 9

Laying hen preference for different pecking blocks in floor pens depends on strain and time of day

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Pecking blocks (PB) are edible enrichment provided to poultry to create foraging opportunities and choice for nutrients. PB vary in chemical composition, texture, colour, and density. This study investigated relative preferences between 3 commercially available PB: A. fiber and mineral, B. fiber, mineral, and molasses, and C. mineral-based. Dekalb White (n=120) and Bovans Brown (n=120) hens were housed in groups of 20 in 6 floor pens per strain. At 19 weeks of age, each pen was provided the 3 PB simultaneously on the litter for 21 weeks with location of each PB rotating weekly. PB were weighed weekly and PB disappearance (PBD) calculated as g/hen/day. PBD was analyzed with a repeated measures GLMM, where fixed effects were strain, PB type, location, and their interactions and pen as a random effect. Pecking behaviour directed at each PB was videoed and quantified weekly for 10 minutes of continuous observation during three time periods: morning, afternoon, and before lights out. Data were analyzed with a binary model to determine whether the PB were pecked (yes/no), followed by a repeated measures GLMM for each strain to compare number of pecks directed to each PB, where fixed effects were PB type, time of day, location and their interactions and pen as a random effect. PBD was affected by an interaction of strain by PB type ($P<0.0001$) with the highest PBD of B for Bovans and C for Dekalb. The type of PB pecked varied at different times of the day for strains, with Dekalb preferentially using C before lights out ($P<0.0001$). Results show a preference for PB type which varies with strain and time of day, providing evidence that the PB may be used to satisfy different motivations that change throughout the day.

Do Mineral Pecking Blocks Support Laying Hens' Ca Appetite?

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Calcium (Ca)-specific appetite refers to the widely recognized behaviour of hens actively seeking Ca-containing food. Individual hens vary in their expression of Ca appetite, yet it is unclear whether this variation addresses specific physiological needs, such as keel bone fractures (KBF) or egg production. This study aimed to examine the association of individual hens' voluntary Ca intake and their physiology. We provided two types of mineral pecking blocks (PB) with different ratios of Ca to Phosphorous (10:1 vs 20:1) to 42 hens of two genetic strains, Dekalb White and Bovans Brown, during an acclimatization period (A: 52-54 weeks of age [WoA]) and one of two trial phases (P1:59-61 or P2:62-64 WoA). The PB disappearance (PBd) was measured at 54, 60, 61, 63 and 64 WoA and feed disappearance (fd), egg production, and KBF severity via radiographs were measured at 61 and 64 WoA. Data were statistically analyzed using R with Kendall's coefficient of concordance (Wt) to assess PB preference stability between A and P1/P2 (long-term) and across the trial weeks (short-term). Total PBd was analyzed with a linear regression model with KBF severity, strain, egg production, fd, and all possible strain interactions as fixed effects. Individual preference for PB types exhibited inconsistency over time (long-term: Wt=0.47, p=0.6, short-term: Wt=0.59, p=0.2). Additionally, PBd rose from a daily mean of 1.62g at 61 WoA to 2.79g at 64 WoA (p=0.006). PB preference appeared unstable, while PBd, a proxy for PB intake, remained unaffected by the physiological state of hens beyond their heightened Ca requirements with advancing age. While our data does not directly support adjusted diet selection, the provision of PBs could mitigate production and welfare issues in older laying hens.

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Theatre 11

Effects of reduced energy and essential amino acid diets on plumage damage and skin injuries of male slow- (Auburn) and fast- (B.U.T. 6) growing turkey strains

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Dietary energy and essential amino acid (EAA) reduction on plumage damage and pecking injuries of male slow- (Auburn) and fast- (B.U.T. 6) growing turkeys were studied. A total of 240 animals were raised in 24 floor pens of 10 animals (12 for Auburn, 12 for B.U.T. 6) over 5 feeding phases of 4 weeks. Three feeding strategies with varying levels of EAA, focusing on lysine and methionine, were tested. The EAA to energy ratio was reduced by 10% to 30% during rearing (until week 8) and up to 10% during fattening (week 9 to 20) compared to breeder company recommendations. Energy content was equal between diets and reduced by 10% per feeding phase. Integuments were scored at week 4, 8 and 20. A multivariate analysis using binary logistic regression models was used to assess the effects of genotype, feeding, age and the interaction of feeding x age. B.U.T. 6 had a higher prevalence of plumage soiling, tail feathers alteration and skin injuries in the back and wings regions compared to Auburn turkeys (P<0.001). Skin injuries, plumage soiling and plumage damage increased with age (P<0.001), with the exception of tail feathers alteration (P=0.357). Feeding (P=0.033) and feeding x age (P=0.014) affected wing condition, with a higher EAA reduction leading to more severe alterations. In conclusion, a more restrictive EAA supply did not result in an increase in plumage damage and skin injuries associated with behavioral disorders, but affected wing condition.

Polyphenols influences neurotransmitter associated with well-being and allometric growth of broiler chickens

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This study assessed the effect of a blend of polyphenols (Silvafeed ATX® – Itália) levels on well-being and allometric growth of broiler chickens. 1,280 one-day-old male chicks (Ross AP91) were housed in floor pens (40 chickens/pen; 8 pens/treatment) in a randomized experimental design with 4 treatments: Control, 250, 500, 1,000 g/t of Silvafeed ATX®. Birds were fed a corn and soybean meal-based pre-starter (1–7 d), starter (8–21d), grower (22–35 d) and finisher (36–42 d). Allometric equations were determined from the weight of commercial cuts of broilers slaughtered at 42 days. On day 40 broilers were submitted to 3 well-being tests: Grab test, modified touch test and latency-to-lie and blood was collected to measure serotonin levels (5-HTT). Student t test was performed to allometric growth. Well-being tests were estimated by the SAS GLIMMIX. Serotonin levels were analyzed using the SAS MIXED procedure and estimates were subjected to regression analysis. It is highlighted that inclusion of 500 g/t promoted late growth ($b > 1$) of wings and breast and early growth ($b < 1$) of back and legs. While breast, legs achieved early growth in broilers fed 250 and 1,000 g/t. Well-being tests did not demonstrate a statistically difference. The level of 5-HTT was increased by the inclusion of the additive reached a better point at 649 g/t. In conclusion, Silvafeed® ATX promoted increase in serotonin levels and modulate the allometric growth of breast muscle growth without changes the final carcass yield.

Session 63

Poster 13

Effects of using a sound signal as a call to feeding on the behavior and welfare of gestating sows

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This study investigates the potential of teaching gestating sows, housed in groups, to feed when an individual sound signal is emitted, in order to reduce aggressions around the feeder. In a test room, 16 “learning” sows went through four individual learning phases (15 min per day for 27 days) to associate an individual sound signal (ISS) as an invitation to feed and to discriminate it from other unknown sound signals (USS). It was followed by a group evaluation phase (1 h 30 per day for 3 days, by groups of 4 sows). Sixteen “naïve” sows were introduced individually to the test room (18 days), and in groups of 4 (3 days) without following the learning procedure. The number of presentations to the feeder following the emission of their ISS (success) and USS (failure), the latency to arrive at the feeder after a sound, and the number of aggressive behaviors were recorded. Generalized linear mixed-effects models were applied to determine the effects of day, treatment (learning, naïve), and sound type (ISS, USS) on the variables measured. Learning sows rapidly associated the sound signal with feed access as they responded to 100% of their ISS after 3 days of training. Distinguishing between different sounds was harder as shown by a percentage of success after an USS emission of 18.8% at the end of learning phases. The latency to arrive at the feeder after an USS increased over days ($P < 0.001$), suggesting that sows could partially discriminate the different sounds. Learning sows were less aggressive than naïve sows on the second day of group phase (7.8 vs 11.7 aggressions per day, $P < 0.05$). This study supports the idea that sows can identify an individual sound signal as a call to feed. The learning strategy reduced the number of aggressions at the feeder, indicating that it could enhance sows welfare.

Effect of a flavouring additive on cow performance in automatic milking system

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Automatic milking system (AMS) requires cows to be self-motivated to enter the robot box. Improving feed palatability consumed in the robot allows to optimize milking frequency. The objective of this study was to determine if feeding a sensory feed additive (SFA; derived from aniseed) improves the number of milkings of dairy cows in AMS. Multiparous mid-lactation dairy cows (160 DIM) were enrolled in this study and housed on six commercial facilities where they were assigned to 1 of 2 treatment diets: 1) control PMR + robot pelleted concentrate (CTRL; n=341), or 2) control PMR with 2 kg of aniseed extract per ton of robot pelleted concentrate (Cristalfeed; Phodé, Terssac, France) (SFA; n=341). Three different types of fodder were equitably represented among diets on those farms: hay, hay + ear corn silage and corn silage. The experimental design included 3 successive periods of 40 days each. Cows received SFA-concentrate during the second period and the control concentrate during the first and third periods. Milking frequency, milk yield and milk composition were recorded daily and analysed by ANOVA with treatment and diet as fixed factors and days in milk as covariate (General Linear Model, JAMOVI 2.3.26). Milkings increased by 3.9% independently of the diet (SFA 2.64 vs CTRL 2.54/d, P<0.001). Milking frequency was positively associated to milk yield (P<0.001) which was higher compared to control (SFA 24.6 vs CTRL 24.1 kg/d, P<0.01). Milk fat increased by 2.7% with SFA-containing concentrate (SFA 4.02 vs CTRL 3.93% (P<0.001) and so was energy corrected milk (SFA 28.7 vs CTRL 27.9 kg/d, P<0.001). The results indicate that the use of an aniseed extract in the concentrate consumed in the robot may encourage cows' behaviour and performance in AMS.

Session 63

Poster 15

Comparative Study of Ruminant Feeding: Nutritional Implications

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This study provides a comprehensive and comparative analysis of ruminant feeding practices, investigating both conventional and unconventional approaches. Within the group of conventional feeds, species such as grass, legumes, mixed grazing, hay grass, corn silage, and alfalfa silage were examined. In the context of unconventional practices, *O. ficus-indica*, *A. americana* L., *A. melanoxylon*, *P. undulatum*, *R. communis*, *A. donax* and *H. gardnerianum* were studied. The parameters evaluated included chemical composition, metabolizable energy, in vitro gas production kinetics parameters, and in vitro dry matter and organic matter digestibility for each species. The results showed significant variations between the forages studied in terms of dry matter, crude protein and different fiber fractions. The significant difference in the dry matter content of *O. ficus-indica* stands out, registering 6.65% when compared to the other species under study. Similarly, the crude protein content of *R. communis*, reaching 24.96% DM, showed notable distinctions compared to the other species, while *H. gardnerianum* showed a significant disparity in its NDF content, registering 72.80% DM. After 96 hours of incubation, *A. melanoxylon* and *H. gardnerianum* showed, on average, lower gas production in vitro. Corn silage stood out as the species with the highest gas production, reaching 56.02 mL/200 mg DM, showing a significant difference from the other plants. *P. undulatum* stood out as the species with the longest lag time, totaling 5.30 hours. These findings outline nuances in fermentation and composition, deepening understanding of nutritional implications for ruminants.

Colostrum administration quickness and farmer's characteristics affect calves' survival

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Colostrum (COL) quality and administration is crucial in dairy farms to guarantee young stock survival. Yet, little is known about the effect of management practices including staff characteristics on calves' mortality. We collected detailed information in 385 Italian dairy herds by interviewing farmers. Mortality rate (%) was calculated as the ratio of n. dead calves in the last 12 mo to the average n. lactating cows, separately for females (MRF) and males (MRM). The ANOVA considered fixed effects of farmer's age (6 class), gender of calves caregiver, and average quickness in COL administration (≤ 2 or > 2 h from birth). Farms (7) leaving calves with the dam in the first days were excluded; within these, the maximum MRF and MRM was 4.00 and 20.00%. In the remaining 378 farms, the maximum declared was 24.63 (MRF) and 13.33% (MRM). Mortality rates were log₁₀-transformed before the statistical analysis to achieve normality of distribution. MRF was significantly affected by quickness, but not by farmer's age. A tendency was observed for the gender of caregiver ($P < 0.10$). In fact, mortality was lower in females managed/inspected by women and taking COL very soon (≤ 2 h). Numerically, farmers over 65 had the greatest/worse MRF. Age of farmer significantly affected MRM, and there was a tendency for the other effects. Calves' survival was worse in farmers over 65 and better when receiving COL ≤ 2 h from birth and managed/inspected by women. Results corroborate the hypothesis that COL management, calves' cares, and staff can affect neonatal survival. On the other hand, factors affecting mortality may differ between males and females.

Session 63

Poster 17

Supplemental rumen-protected dry grape extract alters the metabolic response and rumen fermentation of Simmental cows during heat stress

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Heat stress is well known challenge that negatively influences feeding behavior and metabolism with consequences on health of dairy cows. The aim of this study was to evaluate the effects of supplying a rumen protected dry grape extract (NorGrape® BPO) enriched in polyphenols on metabolic response and rumen fermentation in mid-lactating Simmental cows under natural heat stress. Thirty cows (15/group) were blocked by DIM, MY, and parity and randomly assigned to control (CTR) or treated (BPO) group. In the BPO group, 470 mg/d of NorGrape was mixed with corn meal and top-dressed onto TMR for each cow for 35 d during summer. Blood and rumen fluid samples were collected at 0, 17, 31, and 35 d. Data were analyzed with the PROC GLIMMIX of SAS. Overall, BPO group had greater levels of plasma β -hydroxybutyrate, especially at 17 d, paraoxonase, alkaline phosphatase, and Ca than CTR ($P < 0.05$). Plasma bilirubin and creatinine were, instead, lower in BPO than CTR cows ($P < 0.05$). Regarding rumen, overall, BPO had lower rumen pH and greater levels of total VFA, isovalerate, hexanoate and heptanoate than CTR group ($P < 0.05$). At d 17, D-lactate was lower in BPO than CTR cows ($P < 0.05$). Grape extract rich in water-soluble polyphenols can potentially enhance liver functionality and positively influence rumen metabolism, improving the rumen fermentation efficiency and the rumen environment as reflective of a reduced stressed state, which in turn improve the feeding behavior.

Evaluation of conventional and cow-calf bonded rearing systems in southern Germany

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In traditional husbandry systems, calves are separated from their mothers within 24 hours after birth and bottle-fed with colostrum, following by milk or milk replacer after a few days. An alternative are cow-calf bonded rearing systems, which enable the natural connection between cow and suckling calf. These systems allow the calves, to drink from the udder of their own mother or a foster cow for a longer period (2-30 weeks) after birth. The aim of this study was to carry out a quantitative survey to compare experiences of farmers between two husbandry systems. The survey was completed by 130 farmers and includes questions about personal information, farm characteristics, calf management and animal performance. The first descriptive results showed clear differences in the reported colostrum supply and the separation process management. Cow-bonded calves had faster access to water and roughage and exhibited different behaviors after separation from the mother e.g. vocalization. Comparing statements from farmers with different calf rearing systems helped to identify challenges farmers facing when switching to the cow-bound systems and provide the basis for future experimental research.

Session 64

Theatre 1

Performances of primiparous rabbit does subjected of dietary restriction during gestation

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The ad libitum feeding of rabbit does, especially in the first production cycle, can lead to obesity and may cause reproductive problems and increase mortality in the first parturition. Dietary restriction during certain periods of does' development can be advantageous, reducing this problem and saving on feeding costs. In this study, dietary restriction (approximately 75%) was applied to two groups of nulliparous does between the 4th and 25th day of gestation. During the week, the 30 animals in the R7D group received 136g/day of concentrated feed for 7 days, while the 30 animals in the R6J group received 159g/day for 6 days with one day of fasting. Considering the physiological state and management of the production cycle, litter weight was measured at birth, 11 and 20 days postpartum, and at weaning (34 days of age), and also mortality and individual rabbit weight. Introducing a weekly fasting day significantly increased fertility from 86% to 100%. Rabbit live weight at weaning was significantly ($P=0.045$) higher in the fasting day treatment (more 41 g), but litter weight did not differ. Also, the live weight of the pups and litter did not differ on other considered dates. Gestation time showed a tendency ($P=0.052$) to decrease (by approximately 7 hours), while prolificacy increased ($P=0.07$), from 11.6 to 13.0 total births. The mortality of the pups during the suckling period did not differ and remained around 8.5%, but it's 3,7 percentual points higher in the group R6J. Implementing a fasting day during the gestation period in nulliparous does improve fertility and does not change the other parameters Funding: This work was also supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Effects of dietary valerate glycerides on diarrhea, inflammatory status, and intestinal barrier integrity in weanling piglets infected with Enterotoxigenic *Escherichia coli*

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Two experiments were conducted testing the effects of dietary valerate glycerides on health of weanling piglets infected with Enterotoxigenic *Escherichia coli*. In both experiments, daily diarrhea scores were recorded to monitor the frequency of diarrhea. Serum cytokines were analyzed by ELISA, and intestinal gene expression was analyzed by RT-qPCR. The pilot experiment utilized twenty weanling piglets assigned to control diet, or diet supplemented with 0.1% butyrate glycerides or 0.1% valerate glycerides. Herein, we developed a coinfection disease model, administering two strains of ETEC (F4 and F18; 0.5×10^9 CFU/1.5 mL/strain). Diarrhea frequency was significantly ($P < 0.05$) decreased in either treatment group compared with control. Pigs fed valerate glycerides showed significantly ($P < 0.05$) reduced serum TNF- α on d 4 post-inoculation (PI). Moving forward, a full-scale experiment was performed using sixty weanling piglets challenged with 10^{10} CFU/3 mL F18 ETEC. Treatments included control diet, 0.075% or 0.1% monovalerin, or 0.1% trivalerin. Pigs fed 0.1% trivalerin exhibited significantly ($P < 0.05$) reduced frequency of diarrhea, fecal shedding of ETEC F18, and serum TNF- α compared with pigs in control. The mRNA expression of TNFA in ileal mucosa from pigs fed 0.1% monovalerin was reduced ($P < 0.05$) compared with control. ZO1 mRNA expression was upregulated ($P < 0.05$) in jejunal mucosa from pigs fed 0.1% trivalerin. In conclusion, these experiments provide evidence supporting the capacity of valerate glycerides to reduce diarrhea and systemic inflammation in weanling pigs experiencing ETEC-associated diarrhea.

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Theatre 3

Parabacteroides distasonis alleviates intestinal barrier injury induced by ETEC in piglets

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Enterotoxigenic *Escherichia coli* (ETEC) is the main pathogen causing bacterial diarrhea in piglets. Ningxiang pig has characteristics of stress resistance and low diarrhea rate, which is closely related to the intestinal microbial structure shaped by its high fiber feed. Our previous study found that *Parabacteroides distasonis* (PBd), a representative intestinal bacterial species in Ningxiang piglets, had a significant positive correlation with fecal sIgA. PBd strains were isolated from Ningxiang piglet and weaned mice to test the effect of PBd on intestinal barrier function. PBd significantly increased the sIgA, D-lactate and diamine oxidase (DAO) levels in serum, and up-regulated the muc2, occludin, lgr5 and egf gene expressions in small intestine. We further verified the regulation of PBd on the intestinal injury induced by ETEC in piglets. Our results showed PBd inhibited the colonization of ETEC in intestine mucosa, and decreased the ileal mucosal IL-1 β , IL-4, IL-6, TNF α , IL-10, GM-CSF, TGF β -2 and IL-12 levels after ETEC infection. PBd also decreased the serum DAO activity and D-lactate concentrations, but increased the caludin-1, PCNA and LGR5 protein abundances in ileal mucosa. PBd treatment increased the concentration of acetate and valerate, and affected the structure of intestinal flora in ileal mucosa. Taken together, Ningxiang pig-derived PBd strain could maintain the integrity of intestinal epithelial barrier, alleviate intestinal inflammation and repair intestinal barrier damage caused by ETEC, which provides theoretical basis for nutritional intervention of diarrhea in piglets.

Intrauterine growth restriction, defined by a high brain-to-liver weight ratio, affects fecal microbiota composition and, to a minimal extent, plasma metabolomics in pigs

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Intrauterine growth restriction (IUGR) affects 20% of pigs, altering metabolism and intestinal development. IUGR fetuses redirect blood flow to the brain to survive when nutrient supply through the placenta is limited. This mechanism results in a high brain-to-liver weight ratio (BrW/LW) at birth. This study explored IUGR's effects on fecal microbiota and blood metabolomics. One day after birth, computed tomography was performed on the piglets to assess their BrW/LW. Pigs with the highest (IUGR=12) and the lowest (NORM=12) BrW/LW were selected to collect feces and blood during lactation (T1) and at the end of the starter period (T2), and feces at the beginning (T3) and end of the fattening period (T4). IUGR affected Beta diversity at T1 ($P<0.01$), T2 ($P=0.08$), and T3 ($P=0.03$). At T1, IUGR pigs had higher *Clostridium sensu stricto 1* ($P=0.03$) and *Romboutsia* ($P=0.05$), while NORM pigs showed increased *Ruminococcus* ($P=0.01$). At T2, IUGR pigs exhibited a higher abundance of *Prevotellaceae NK3B31* group ($P=0.02$), *Rikenellaceae RC9* gut group ($P=0.03$), and *Alloprevotella* ($P=0.03$), while NORM pigs had increased *HT002* ($P=0.05$). At T3, IUGR pigs had higher *p-2534-18B5* gut group ($P=0.03$), while NORM pigs showed an increase of *Prevotella_9* ($P<0.01$). None of the plasma metabolites was significantly different at T1. However, at T2, asparagine was lower in IUGR compared to NORM pigs ($P<0.05$). Our study demonstrated that IUGR affects fecal microbiota composition until the beginning of the fattening period. By contrast, the blood metabolome was minimally affected.

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Theatre 5

Impact of moderate CP restriction and additive combination on N balance and hindgut microbiota in pigs

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The aim of this study was to compare different dietary treatments on nitrogen (N) balance, hindgut microbiota and fecal excretion of microbial biomass in grower-finisher pigs. A 2 x 2 factorial design was conducted with 32 male non castrated pigs fed in a 2-phase production cycle: grower (G; 68.4 ± 2.18 kg BW) and finisher (F; 108.8 ± 1.30 kg BW) with 2 crude protein (CP) levels (SP: 16.5 and 15% CP vs LP: 15 and 13.5%CP), and without (CTR) or with a mixture of additives (A; carbohydrases, organic acids, and essential oils). Four pigs/treatment and phase were allocated in metabolic cages for individual urine and feces collection to determine N balance and total tract digestibility. Animals were sacrificed and ileal and colon digesta were analyzed for nutrient digestibility and microbiota characterization, respectively. Fecal microbial N load, DAPA: N was analyzed by UPLC-MS. Statistical analysis was performed using JMP®, with GLM procedures and Tukey's test, and multivariate analysis from microbial data was conducted in R (v 3.6.3). N net retention was higher in LP diets and specially in F phase ($P<0.05$). Finishers had higher ileal digestibility (DM, OM and CP), but no differences were observed between treatments, and F showed a more diverse colon microbiota. LP reduced alpha diversity and microbial load in CTR animals, and the inclusion of additives led to an overall increase in alpha diversity and microbial load (μg DAPA per g DM, $P<0.001$) promoting hindgut maturation in the pigs. To conclude, finisher animals fed with LP diets were more efficient in terms of N retention but it compromised hindgut microbial diversity, impact which was controlled with the supplementation of a mixture of feed additives.

Microbial architecture and enterotypes for improved nitrogen utilization efficiency in fattening pigs

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As global demand for pork continues to rise, strategies to improve nitrogen utilization efficiency (NUE) in pig farming are mandatory for environmental sustainability. This study explored the interplay between fecal microbiota, volatile fatty acids (VFAs) and NUE in pigs with a defined family structure. Pigs (n = 508) were kept under standardized conditions and a two-phase feeding regime with marginal lysine supply (90% of recommended). Fecal samples were collected from all pigs in both phases (SP) and analyzed for VFA content and microbiota composition. In each SP, pigs were assigned to two groups based on <25%- and >75%-quantile of NUE. Our findings revealed significant effects of sire, SP and sex on fecal microbiota composition. LinDA model identified *Streptococcus* as a potential biomarker for NUE in SP1. The offspring of some sires were either entirely absent or unevenly represented in the groups, suggesting a genetic predisposition to NUE regarding the fecal microbiota. The identification of two enterotype-like clusters, *Lactobacillus* (LACTO) and *Clostridium sensu stricto* (CSST), further underscored the microbiota role in NUE. Pigs' affiliation with enterotype-like clusters altered over time and might be sex-dependent. The cluster CSST correlated with higher VFA concentrations and NUE, but lower daily nitrogen retention, suggesting enterotype-specific metabolic efficiencies. This research shows the microbiome's potential to modify NUE, paving the way for future strategies to improve sustainable pig production.

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Theatre 7

Effect of dietary inclusion of a herbal extract blend on nutrient digestibility, faecal Nitrogen emission and growth performance in growing-finishing pigs

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The improvement of nutrients absorption may be a strategy to increase the productive, economical and environmental efficiency of pork production. The objective of this study was to evaluate the influence of the dietary inclusion of a herbal extract blend (q-Cebo, Ecovet Economía Veterinaria S.L.) based on chicory inulin (dried roots of *Cichorium Intybus*), inactivated yeast fermentation product (*Saccharomyces cerevisiae*) and *Yucca Schidigera* juice on nutrient digestibility and growth performance in growing-finishing pigs. A total of 132 [Pietrain x (Landrace x Large White)] pigs (n = 11 replicates, 39.5 ± 3.42 kg body weight, BW) were used. Half of the pigs were fed with a common basal diet (Control group), while the other half were fed with a similar nutritional diet with 1 kg of q-Cebo/Tn of feed supplemented, considering its nutritional matrix (q-Cebo group). Growth performance and total tract apparent retention (TTAR) of ash, crude protein (CP) and ether extract (EE), and faecal Nitrogen excretion (N) were calculated. The study lasted 69 days with a final BW of 111.6 and 111.7 kg, control and q-Cebo pigs, respectively (p > 0.1). The inclusion of q-Cebo did not influence any performance parameter (p > 0.1). Whereas pigs fed with q-Cebo had greater TTAR of CP (88.7 vs. 90.8%), EE (80.9 vs. 85.3%) and ash (68.4 vs. 72.8%), and a lower N (421.6 vs. 351.3 g N/pig) than Control pigs (p < 0.001). In conclusion, q-Cebo dietary inclusion considering its nutritional matrix is a promising strategy to increase nutrient digestibility and to reduce environmental N, while maintaining growth performance in growing-finishing pigs.

From gut to tail: The effect of a fermented herbal extract on aspects of health and (tail-in-mouth) behaviour of pigs

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In pig farming, respiratory problems and tail biting are frequent challenges. Adding direct-fed microbials to the feed might not only be beneficial for the health of an animal but also influence its behaviour through the gut-brain axis. This study investigated the effect of a fermented herbal extract (FHE) which included a blend of herbs, lactobacilli and yeasts. All animals were examined from weaning to slaughter (n=760) on 3 commercial Austrian farms which are part of a welfare label and raise pigs with intact tails. The goal was to determine if FHE is effective in improving aspects of animal health (respiratory diseases, microbiome composition) and welfare (tail injuries and length). Fecal samples were collected to assess the microbiome composition (16S rRNA amplicon sequencing). Using mostly generalized linear mixed models, almost no significant differences were found regarding behaviour and health status. However, at the end of the finishing period, more FHE animals remained with normal tail lengths compared to controls (p=0.009). FHE pigs also coughed and sneezed less during behavioral observations on farm (p=0.026). Concerning microbiome composition, farm and age had a bigger impact on the fecal bacterial communities than treatment (both p=0.001). Although inconsistently across farms, FHE promoted microbiome diversity in rearing piglets. While FHE cannot be the sole measure to solve tail-biting in pigs, it can support raising more pigs with intact tails.

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Theatre 9

Vitamin E reduce adipose tissue fibrosis and lipid metabolism disorder in oxidized fish oil-fed pigs

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The research explored how oxidized fish oil impacts the growth, lipid metabolism, and adipose tissue health in weaned piglets, and whether antioxidants could counteract these effects. Forty piglets were assigned to four dietary groups: CON (basic diet + 2% fresh fish oil), OFO (basic diet + 8% oxidized fish oil), VE (OFO diet + added vitamin E), and TBHQ (OFO diet + added tert-butylhydroquinone). The findings were clear: The OFO group experienced hindered growth, inferior carcass quality, and negative changes in lipid profiles compared to the CON group. The VE and TBHQ groups showed improvements in average daily gain (ADG) and average daily feed intake (ADFI), along with better feed-to-gain ratios (F/G) relative to the OFO group. The VE group, in particular, demonstrated enhancements in carcass weight and dimensions, serum lipid levels, adipocyte size, and a favorable shift in fatty acid composition. This group also showed increased gene expression related to fat breakdown. The OFO group exhibited signs of adipose tissue fibrosis, which was not seen in the CON group. This fibrosis was lessened in the VE group. In summary, while oxidized fish oil had detrimental effects on the piglets' growth and metabolism, dietary antioxidants like vitamin E were effective in mitigating these negative outcomes.

Effect of oat milling (ground vs. flaked) and age on the fecal microbiome of piglets

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This study aimed to determine the effect of grinding oats on the faecal microbiome of piglets. The trial involved 2 experimental treatments: ground oat (GO) and flaked oat (FO). The feed was distributed from 7 days of age, still in the maternity, until the end of the experiment. Weaning occurred at 28 days, and 36 animals were divided into 12 pens with 3 animals each. 24 animals were slaughtered at 32 (Age 1) and 39 days (Age 2) of age (12 at each age and in each treatment). In slaughtered animals, fresh faecal samples were collected. The faecal microbiome was characterized using the 16S rRNA gene and shotgun metagenomic sequencing. The Wilcoxon signed-rank test revealed no significant differences in alpha diversity across all evaluated experimental groups, irrespective of age, as well as within each age group across the experimental conditions. We also observed that the effect of the age explains around 14% of the variability of the beta diversity among groups ($p < 0.001$). Within each age, the effect of the diet in Age 1 accounts for around 12% of the observed variability. No significant differences were observed between experimental groups within Age 2. Thus, how oat are included in the diet influences the microbiome's diversity in younger animals (32 days)

Session 64

Poster 11

Effects of vinegar intake on piglet performances and gut health

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The objective of the work was to evaluate the effect of vinegar added to drinking water on the productive parameters and fecal microbiota of piglets. The study involved 24 piglets distributed in 3 pens (8 piglets/each), and 3 treatments: drinking water (CT), drinking water with 2% of wine vinegar (VV) or cider vinegar (VS). All groups received a commercial diet ad libitum. Live weight was measured and gastrointestinal disorders (diarrhea) were assessed for 4 weeks starting at weaning (35 days). At 63 days of age, fecal samples were collected to determine the intestinal microbiota. No significant effects were observed in growth (final live weight of 22.21 kg and average daily gain of 470 g/day) and in the occurrence of diarrhea. The cider vinegar promotes a significant ($P < 0.05$) reduction in the population of *E. coli* and lactic acid bacteria compared to the CT treatment but does not affect total coliforms. The wine vinegar only reduced the lactic acid bacteria count. The vinegar reduced the concentration of some types of microorganisms but did not affect the growth performances of the piglets. It will be important in future work to increase the replicas and other concentrations of vinegar

Effects of 20% Lactose Supplementation in Liquid Creep Feed on Piglet Growth

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The initial weeks of a piglet's life are crucial, with challenges such as diarrhea, decreased growth, and reduced feed intake, particularly around weaning. Previous research suggests that adding lactose to piglet diets can enhance growth and health by increasing feed intake, thereby improving resilience at weaning. This study aimed to evaluate the effects of a liquid creep feed containing 20% lactose versus a control with 0% lactose on piglet growth from days 10 to 28. Twenty-seven piglets were individually housed and fed a milk replacer (MR) from day 1 to 10. From days 10 to 28, piglets were allocated to one of three treatment groups: control (CON), which continued receiving MR; MR and liquid feed (MRLF); and MR and liquid feed containing 20% lactose (LAC). Body weight and feed intake were monitored daily. After the end of the experimental period, all piglets were euthanized, and blood and SI samples collected for later assessment of hormone and gene expression levels. The analysis was conducted using the ANOVA function in R, using a linear mixed model with treatment and sex as fixed effects. Contrary to expectations, in the last part of the growth period, the CON group had greater body weight compared to the LAC group ($P=0.001$). Piglets in the LAC group had lower final body weight compared to the control group, even though there was no significant difference in average daily weight between the groups. This finding suggests that the strategy of adding 20% lactose to the diets did not enhance growth performance in the expected manner. Despite the lack of effect on phenotypical growth parameters, further analysis might reveal alterations in gut function or appetite regulation.

Session 64

Poster 13

The effect of Benzoic Acid supplementation on growth performance and small intestine integrity of artificially reared suckling piglets

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Reducing the severity of diarrhea in the peri-weaning period is of great importance, considering its negative impact on piglet's health and growth performance. The small intestine (SI) of the pre-weaned piglet is immature, and thus highly susceptible to infection, causing diarrhea. Benzoic acid (BA) has been shown to improve integrity and function of the SI in post-weaned piglets, but its potential effect has not been investigated in suckling piglets. The aim was to study the effect of BA supplementation on growth performance and SI integrity and function, to improve robustness of piglets prior to weaning. The study included 27 individually housed 1-day old piglets ($BW=1195g \pm 152g$) equally divided in three dietary treatments: Milk replacer (M), M+liquid creep feed (MLF) and M+LF+0.5% BA (MLFBA). In the first period ($P1=d 0-10$), piglets from all three dietary treatments only received milk replacer; in the second period ($P2=d 10-28$), piglets from MLF and MLFBA were in addition provided with liquid creep feed without or with BA, respectively. Dry matter intake (DMI) and BW were registered daily, and ADDMI and FCR subsequently calculated for. At the end of the trial, all piglets were euthanized, and SI samples were collected for later assessment of morphology characteristics and levels of gene expression. The data were analyzed using the ANOVA function in R with a linear mixed model including treatment and sex as fixed effects, and litter as random effect. No differences in BW, ADG, ADDMI or FCR between dietary treatments in the overall period were found. In P2, MLFBA displayed significantly greater ($p < 0.05$) ADFI compared with MR. In conclusion, BA did not improve growth performance of artificially reared suckling piglets.

Computational and Experimental Framework for Tracking Cow Personalities and Their Impact on Personalized Welfare

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Understanding animal personality can improve animal welfare, management practices, and advance our biological understanding of individual differences. Building upon our previous research, which introduced a mathematical framework for characterizing animal personality traits, we have extended this methodology to encompass the study of dairy cows. To achieve this, we developed a system to continuously track the positional data of each cow within a 70-cow herd over several months. This enabled the automatic computation and construction of a multidimensional behavioral space encompassing over 100 behaviors for each cow on a daily basis. We identified personality traits underlying behavior, showing inter-individual variation and intra-individual consistency over time, persisting beyond a year. These traits correlate closely with behavioral and physiological parameters like dominance rank and milk production, offering insight into herd dynamics and productivity. Our approach supports “personalized welfare” in husbandry, tailoring interventions to individual cows’ unique behavioral profiles. Detecting subtle changes in behavior over time aids early identification of health and welfare issues, enabling timely interventions. Through this longitudinal study, we demonstrate the framework’s versatility beyond labs, promising widespread application in animal sciences. It provides a precise means of measuring personality, illuminating the interplay between behavior, affective states, physiology, and productivity in animals.

Session 65

Theatre 2

Validation of an indoor positioning system for automated personality assessment in dairy cows

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Abnormal behaviors serve as indicator for sickness, injuries, and stress. Since behavior is guided by personality, abnormal behavior can differ between herd members with different personalities. Gold standard for personality assessment are behavioral tests which are time-consuming and hardly cover interactions between different personalities within the herd. To automatically collect behavioral data, including social interactions of cows, we installed an ultra-wideband positioning system at the research farm of Kiel University. The system is able to triangulate beacons in a two-dimensional space once per second. Exact positions are calculated through a time difference of arrival algorithm in an energy saving way. To identify social contacts between individual cows a high accuracy of the calculations is needed. Evaluation of tracking accuracy was done through tracking two stationary beacons for a period of one hour and equipping a human with a beacon. The human walked predefined routes through the stable in a steady pace of one step per second. For stationary beacons 70 % of all possible triangulations were successful with normally distributed residuals around true locations (standard deviation of 26 cm on the x-axis and 22 cm on the y-axis). Within the predefined routes 50 % of all triangulations were successful with accuracy ranging of ± 24 cm on the x-axis and ± 75 cm on the y-axis. In the ongoing of our research, we are planning to equip all cows with a beacon, to investigate links between dairy cow personality, which we established through behavioral tests, and home pen behaviors.

Automatically monitoring dyadic relationships in dairy cows

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Maintaining meaningful social relationships is an important component of welfare, yet monitoring social bonds in dairy cow groups is challenging. This study explores the potential of utilizing data generated at the feed bunk and milking parlor to analyze dyadic relationships among cows. For 10 days, we followed 2 stable groups of 48 lactating cows with access to 30 electronic feeders. Feeder and parlor data was used to measure the frequency of agonistic replacements at the feeder, the duration of feeding neighbor events, and the frequency with which cows were milked next to each other. Separate social networks were created for replacements, feeding neighbors, and milking neighbors and we assessed the association between these. Replacements showed low dyadic reciprocity in both groups, consistent with the notion of dominance relationships, and dyadic replacements were not influenced by the time spent as feeding neighbors. In the two groups, dyads spent on average 4.9 ± 2.6 and 5.2 ± 2.9 min/day as feeder neighbors. Fitting a mixed distribution to this data indicated avoidance and affiliation relationships in both groups, characterized as < 4 min or > 9 min average daily feeding neighbor time. Being neighbors in the milking parlor was weakly associated with being feeding neighbors in one group (regression coefficient: 0.26, $P < 0.001$) but not in the other (regression coefficient: 0.11, $P = 0.1$). It is important to recognize that factors other than social preference may affect feeding and milking behaviors. We suggest evaluating less strict criteria for defining shared resource use to better identify consistent dyadic bonds among cows.

Session 65

Theatre 4

Assessing Dairy Cows' Welfare Status through Social Behavior Indicators Using an Automatic Real-Time On-Farm Positioning System

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The intensification of dairy farming has led to larger, more crowded farms, escalating concerns about animal welfare across the industry. This study aims to relate non-intrusive animal behavior indicators to animal welfare. Elevated cortisol levels, a stress biomarker, were hypothesized to correlate with alterations in social positioning, potentially reflecting health issues. A high-throughput Ultra-Wideband technology was deployed to track the real-time positioning of 70 cows in a commercial dairy farm. Over three weeks, behavioral data were recorded alongside weekly cortisol samplings from 30 randomly selected cows; coinciding with these samplings, observations to assess dominance hierarchies were also carried out. Additionally, we compiled records of milk production, health, insemination, and conception. Continuous centrality metrics, i.e., the individual cow's distance from the herd's center of mass, were computed. Correlations were sought between centrality and factors like dominance, age, cortisol levels, milk, and health. Preliminary results indicated a relationship between increased cortisol and lower centrality, suggesting stress induces avoidance behavior. Notably, shifts in centrality preceded the emergence of diseases, in addition, an association was found between elevated cortisol levels and the incidence of disease. Initial findings suggest that social behavior has the potential to provide insights into cattle welfare, underscoring the need for further research. By establishing innovative welfare benchmarks, farms embracing ethical husbandry will benefit from improved livestock welfare and financial advantages.

Full-flock-full-pregnancy monitoring of liveweight progression in Romane meat ewes, using a walk-over-weighing system in rangelands

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Little is known about liveweight (LW) dynamics of females during pregnancy, which is more critical under rangeland conditions. In this work, we implemented fine monitoring (daily) of the LW progress of each Romane ewe belonging to the La Fage flock, throughout the whole pregnancy, and with the help of our walk-over-weighing (WoW) platform. The trial involved 279 ewes (77 primiparous and 202 multiparous) which initial average age (and LW, kg \pm SEM) were 1.5 (56.5 \pm 1.98) and 4 years, 3 months old (79.3 \pm 1.15), respectively. A period of adaptation (around 2 weeks) was necessary to voluntarily cross the WoW (achieved by 80% of ewes). Raw dataset outliers were automatically filtered by our free web app ORIOLE (<https://oriole.sk8.inrae.fr/>). The final, clean dataset was contrasted with other relevant individual information available e.g. pregnancy diagnosis at mid-pregnancy (~72 \pm 8 d), number of fetus, litter size, sex and LW at lambing. Constant ewes' LW progress was easily followed by the WoW, making possible to detect anomalies e.g. abortions or individual presence/absence at a given precise date. The infrastructure is ideal for making further deep interpretations such as fetus growth rate in function of litter size, lamb' sex, the relationship with LW at lambing etc. Also, for contributing to the development of early warning systems helping to decision makings during such a critical physiological stage in the farm and beyond. This work was in the scope of the H2020 TechCare project (GA 862050).

Tracking sheep indoors or on pasture using Bluetooth and UHF RFID for welfare management: feedback from trials conducted in Scotland and France

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Precision Livestock Farming tools and digital technologies can have value for welfare management, including assessment of behaviour and detection of any changes. It can provide non-diagnostic and non-specific indicators to the farmers, informing them that welfare may have been impacted, and that further investigation is needed. Monitoring the visits to points of interest and/or resources, such as water trough or feeder, can be useful in the potential detection of welfare problems. Bluetooth beacons (BLE beacons) and Ultra High Frequency (UHF) RFID tags have been tested to track sheep' attendance in different environmental conditions, alongside regular welfare assessments. In Scotland, BLE beacons have been used to monitor the attendance of ewes at feeding points, as well as the ewe-lamb bond on pasture. UHF tags have been tested in Scotland to monitor visits to resources on pasture, whilst in France they have been used indoors to monitor the water trough attendance by adult dairy ewes and fattening lambs. Here we will compare the advantages and disadvantages of the use of these technologies in the different trial environments. Although these technologies offer encouraging preliminary results, there are important issues of adaptation to the breeding environment (humidity, metallic environment in the shed, power supply and battery life, etc.), which need to be taken into consideration to assist with the development of future trials using these technologies in a variety of environments and grazing systems.

Milking order of healthy and subclinically mammary infected dairy ewes in mid lactation

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Udder health (mastitis) is a first prioritized issue for dairy sheep welfare and PLF tools offer new opportunities to reduce intramammary infections (IMI). IMI leads to poor welfare (fever, pain...), which compromises behaviour. This study assesses the use of milking order (MO) as a proxy for udder health and milk production. With this aim, 78 Lacaune (LC) and 34 Manchega (MN) dairy ewes identified with low frequency (LF) boluses (22 g, Datamars, SW) were used in mid lactation (d 110 to 140). Milking was done twice daily in a 2x12 milking parlour with MM25SG electronic milk meters and 2 LF reading gates (DeLaval, ES). MO and milk yield (AM and PM), milk composition (monthly), somatic cell count (SCC) by udder half (monthly) and BW (monthly) were recorded. Milk bacterial culture (blood-agar, 37°C for 24-48 h) was done when $SCC > 500 \times 10^3$ to assess IMI (positive ≥ 5 cfu). Statistical analyses were performed using R. Pearson and Spearman correlations were used for quantitative or ordinal variables, respectively, and means compared by T-Student or Wilcoxon Mann-Whitney tests, respectively. Repeatability of MO was measured as intraclass Fisher's correlation. No clinical mastitis was detected during the study, but SCC increased from 300 to 739×10^3 , whereas IMI only increased from 28.6 to 32.1%. Most IMI were CNS (Coagulase negative staph., 33 to 50%) and SA (Staph. aureus, 30 to 17%). No correlation was found between MO and milk yield or $\log_{10}SCC$. On the contrary, correlations with MO were positive for milk fat and protein contents ($Rho = 0.21$ to 0.26 ; $P < 0.01$) and negative with lactose ($Rho = -0.31$ to -0.26 ; $P < 0.01$). BW correlated positively with MO ($Rho = 0.40$ to 0.47 ; $P < 0.001$). Wilcoxon Mann-Whitney correlations of MO showed effects of breed (MN = 70 and LC = 50; $P = 0.016$ to 0.014) and parturition (1st = 32 and >2nd = 65; $P = 0.001$). IMI only correlated with MO at d 140 (Negative = 44 and Positive = 69; $P = 0.021$). In conclusion no evidence of effects of udder health on milking order was detected, although differences by breed, parturition and BW of the ewes were observed. Acknowledgements: UE Program H2020 (Project TechCare #862050)

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Theatre 8

How to define good mothering ability using computer vision of free farrowing sows?

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Most of the sows are housed in a cage system in the farrowing unit. Although this system is intended to reduce preweaning mortality, the welfare of the sow might be compromised as it greatly restricts their natural behavior. Free farrowing systems allow sows to move around in the farrowing pen and this might stimulate nesting behavior, shortened farrowing duration and improved locomotion score and therefore the longevity of the sow. On the other hand, the risk of increased preweaning mortality in free farrowing system, due to crushing, is higher compared to cage systems. Previous studies suggest that sow behavior influence preweaning mortality as some sows are more careful than others, also known as mothering ability. To investigate mothering ability of sows in free farrowing systems, we installed 40 cameras above free farrowing pens to observe the behavior of the sow. We will train a computer vision model (YOLO) to estimate the contour and posture of the sow and the position of the piglets. Using these models, the behavior and the movement pattern of the sow could be monitored on a continuous basis. The dataset is continuously growing and will add data of 416 sows on a yearly basis. Performance of sow and piglets are recorded, for instance sow weight, total number born, number weaned, piglet weights. A first pilot showed the ability of the computer vision algorithm to detect the sow and her posture accurately, and the models are now extended to detect piglet location.. This will allow us to find beneficial behavior of sow and piglets, estimate genetic parameters for potential implementation in breeding programs, and by this improve performance in a free farrowing system.

Improvement of an algorithm performance to estimate the physical activity of group-housed sows based on videos

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In a previous work, a deep-learning algorithm has been developed to automatically detect the different physical activities of the gestating sows on images with an overall accuracy of 66%. However, the classification accuracy varied regarding the activity detected from 23% for sitting to 86% for eating. Therefore, to improve this algorithm a more recent architecture (yolo v8 instead of the initial tensorflow) of the neural network has been constructed and evaluated with the same data sets. Several groups with an average of 18 gestating sows, housed in similar pens but equipped with 2 or 3 cameras each, were recorded continuously during their gestation. Six activities (lying ventrally, lying laterally, sitting, standing, eating and drinking) were manually annotated on 3,317 images extracted from the videos (33,894 sows annotated). A set of 80% of these annotated images was used to train the algorithm, an object detection model that uses convolutional neural networks to detect and classify objects in an image. The second set with 20% of the annotated images was used to validate the performance of the algorithm. With this new algorithm architecture, the accuracy of the algorithm (on average 97%) was strongly improved as well as the accuracies of all the activities detected ranging from 96% for sitting to 99% for standing. In parallel, this algorithm is being evaluated on another set of images recorded by different types of cameras in a new environment, and adjusted to reach similar accuracies. To go further and use this tool to estimate individual welfare, coordinates of the animals are being recorded continuously and interpreted to quantify the number and type (positive vs. negative) of interactions between sows.

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Theatre 10

Computer vision based monitoring of fattening pigs: how well do pigs behave?

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Until now, monitoring of fattening pigs has been focused primarily on production parameters such as feed and water intake and weight. However, camera monitoring using computer vision based approaches can offer new opportunities to continuously monitor both production parameters and animal behaviour. Specifically, monitoring the animal behaviour can enable insight into animal welfare issues such as tail biting, improve the knowledge of fattening pigs social structure in a pen or can even assist in modelling pig growth. The goal of this research is to combine traditional PLF technologies (RFID antennas, flow meters, weight scales) with state-of-the-art computer vision models in order to create the link between growth and behaviour. In this study, 120 pigs (1 compartment, 8 pens) were continuously monitored by an angled top view camera (1 camera per pen). The resulting videos were analyzed using modular computer vision deep learning models for detection (M3det), tracking (M3track), pose estimation (M3PoseSeg) and pig interactive behaviour (M3ethology). Furthermore, all pens were equipped with RFID antennas at the feeder and drinker, water flow was measured at all drinkers, feed intake was measured in 4 pens and pig weight was recorded in each pen by a weight scale at the feeder or drinker. The computer vision tracking was also assisted by detection of large ear tags and by the RFID identifications to improve the length of individual pig tracks. Possible features (such as pig activity index) as a result of the computer vision models were explored and combined with the measured production parameters.

The effect of colostrum supplementation on milk intake and performance when administered at the time of a diarrhea alert

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This randomized clinical trial aimed to evaluate the effects of intervening with colostrum at the time of a diarrhea alert on diarrhea duration, milk intake, and performance in Holstein dairy calves. Eighteen calves were offered 15 L/d of milk replacer by an automated milk feeder where an alert validated to identify calves at risk for diarrhea was programmed ($\leq 60\%$ rolling dividends in milk intake and/or drinking speed over 2 d). At the first alert, calves were randomly allocated to receive either 1 L of colostrum replacer mixed at 130 g/L (CR) or 1 L of milk replacer mixed at 150 g/L (MR) by bottle for 4 consecutive d. Treatments were balanced for metabolizable energy. Fecal consistency was scored daily, and diarrhea was diagnosed when calves had loose feces for ≥ 2 d or watery feces for ≥ 1 d. Body weight was collected at birth, on alert d, and 2x weekly thereafter. Mixed linear models evaluated the association of treatment with milk intake, drinking speed, and ADG for 7 d after the alert, and final body weight at 7 d after the alert. Alert weight and alert age were included as covariates. Calves triggered the alert at (mean \pm SD) 10 ± 3 d of age and were diagnosed with diarrhea at 11 ± 2 d. Duration of diarrhea after the alert was similar between treatments (CR: 3.4 ± 0.63 vs MR: 4.3 ± 0.63 d, $P = 0.34$). There was no association of treatment with milk intake (CR: 8.7 ± 0.62 vs MR: 7.4 ± 0.59 L/d, $P = 0.15$) or drinking speed (CR: 0.40 ± 0.03 vs MR: 0.35 ± 0.03 L/min, $P = 0.31$). Calves provided CR had greater ADG (CR: 1.01 ± 0.11 vs MR: 0.67 ± 0.11 kg/d, $P = 0.04$) and weighed 5 kg more at 7 d after alert (CR: 55.3 ± 1.50 vs MR: 50.5 ± 1.49 kg, $P = 0.01$). Preliminary results from this study suggest that providing colostrum at the time of a diarrhea alert could improve weight gain over the 7 d after the alert.

Session 65

Theatre 12

Redefining lameness assessment using crowd-sourced data

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Lameness is a prevalent issue affecting on average 23% of dairy cows globally. Lameness causes pain to cows and economic losses to farmers, but can be difficult to detect and routinely monitor due in part to the subjective and labor intensive nature of traditional gait scoring methods. Here, we propose a novel approach for assessing cattle lameness, where crowd workers compare video pairs to determine which cow is more lame and the results are leveraged to develop a lameness hierarchy. In the pilot study of videos of 30 cows, we found traditional gait scoring systems to be subjective and inconsistent. Calculated using the intraclass correlation coefficient (ICC), the intra- and inter-observer reliabilities were 0.62 ± 0.09 and 0.44 ± 0.02 , respectively. In contrast, our lameness hierarchy method achieved high inter-observer reliability among experienced assessors (ICC = 0.81), and demonstrated strong agreement between experienced assessors and untrained crowd workers (ICC = 0.85). Using a subsampling algorithm that we created, we found that only 8 crowd workers per video pair are needed to reliably assess lameness, reducing the total number of comparison by 61% compared to using all possible pairwise comparisons. We conclude that our proposed lameness hierarchy method facilitates quick and accurate labeling of lameness videos, which can be used to create large training datasets suitable for developing reliable automatic lameness detection models.

Genetic relatedness impact on social behavior in dairy cows within Automated Milking Systems

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Comprehending social dynamics among dairy cows is essential for optimizing herd welfare and productivity. Our study aims to evaluate the impact of genetic relatedness within a cow herd on their social connections using Social Network Analysis (SNA). We identified consistent social bonds among cows by gauging the time interval between consecutive milking events, i.e., the duration from one cow entering the milking robot to the subsequent cow's entry. If the timeframe between two cows entries at the milking machine is less than 10 minutes, we consider it an association between them, as it is highly likely that they were queuing at the same time. We assessed the genetic relatedness among cows by examining pedigree data spanning three generations of both cows and bulls. Where accessible, the pedigree-based relatedness will be compared with the genomic kinship matrix, to verify if a detailed analysis of the cows' DNA profiles, providing more detailed information about the degree of relatedness among individuals within the herd, will further help understanding social dynamics influenced by genetics. We examined the influence of genetic relatedness on cows' social preferences throughout different lactation periods. Additionally, we investigated the connection between these social bonds and milk yield, fat, and protein content. Finally, we compared individual cows' ego-networks to understand genetic relatedness effects and assess the stability of their social connections over time. This study emphasizes the importance of understanding social behavior in dairy cows, offering crucial insights for enhancing herd management practices in the dairy farming industry.

Automatic detection of lame weaner pigs by evaluating their drinking behaviour

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This study aimed to detect lame pigs by analysing their mean daily water consumption (WC) and number of drinking events (NDE). Therefore, WC and NDE of 714 weaner pigs with RFID-ear tags from five batches were evaluated. Modified drinking stations with RFID-antennas and waterflow meters assigned each pig to its retrieved water amount. Treatments due to lameness were recorded in 38 pigs, and their WC and NDE on the seven days before the first treatment (DBT) were compared to the group mean. Effects of lameness and DBT on the WC and NDE were statistically analysed. Treated pigs had a significantly lower WC than their pen mates in the observation period. The NDE and DBT interacted significantly with a reduced NDE for treated pigs on treatment day, compared to the days before treatment and the group mean on this day. Evaluating WC and NDE revealed different drinking behaviour in lame pigs. Further investigations will focus on the reduced WC of lame pigs and include early clinical symptoms to evaluate the benefits of automated detection compared to human observation. The study is part of the project DigiSchwein, which is supported by funds of the Federal Ministry of Food and Agriculture (BMEL) based on a decision of the Parliament of the Federal Republic of Germany. The Federal Office for Agriculture and Food (BLE) provides coordinating support for digitalisation in agriculture as funding organisation, grant number 28DE109E18.

Requirements for implementing a European knowledge platform for multidimensional animal welfare assessment on farm, during transport and at the slaughterhouse

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The present project aimed to construct a framework for a knowledge platform that enables monitoring and surveillance of animal welfare (AW) indicators at European level on farms, during transport and at the slaughterhouse. This involves the enhancement of the integration, interpretation, and dissemination of AW data, all while adhering to the FAIR (Findable, Accessible, Interoperable, and Reusable) principles. An initial investigation was performed into existing database systems currently utilized throughout the food supply chain, focusing on the platforms in use, their access rights and data management capabilities. This was followed by a concise survey designed to gauge the preferences of the scientific community and stakeholders within the AW domain regarding the desired content and architectural design of the upcoming AW knowledge platform. The expected outcome is a comprehensive set of criteria and compromises essential for creating a reference platform at the European level, which aims to facilitate AW monitoring, surveillance, and benchmarking among participating entities, as well as enhance the exchange of data among various stakeholders. Keywords: Animal welfare, knowledge platform, data integration, requirements, FAIR principles

Session 65

Poster 16

The effect of precision lighting system on laying hen welfare

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A precise, remotely controlled LED lighting system (PLS) for laying hens was developed with the following parameters: white 50-115 + red 50-100, 15-36 lx, 14 hours of light, which was used on a farm scale (hen house with an area of 802 sq. m). Leghorn hens at the age of 25-74 weeks were observed using a monitoring system (IPOX NMVS-2.0). The results obtained in the experimental hen house were compared with retrospective results for the same line of hens in the control house (C) with incandescent lighting (13 lx, 60 W, 2 rows × 17 bulbs). The PLS proved to be more beneficial for hen behaviour. They were better at climbing the grates and were calmer. The number of non-nest eggs was statistically significantly lower. The PLS used did not provoke undesirable behavior in hens. The laying curve of the birds indicates that the hens started laying on time and that the decline in laying was much milder in PLS compared to C hen house. However, the color of light must be correlated with its intensity and color temperature so as not to cause an undesirable reaction of the hens during the period of intensive development. The tested PLS, thanks to a wide range and quick ability to change lighting parameters (without the need to enter the flock) depending on the needs of birds in a given physiological period, proved to be a convenient and effective tool in precise production of eggs. Funded by the Minister of Science under „the Regional Initiative of Excellence Program”.

Engineering resilient grazing systems to improve adaptation to climate change and helping to mitigate its adverse effects

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Anthropogenic processes are driving climate change resulting in increasing temperatures and more frequent extreme weather events such as drought and flooding. It is important to develop strategies not only to adapt to the changing climate, but also to mitigate its impacts in livestock systems. Diversifying the forage species is an important step to enhance livestock system resilience. Diversification might happen both in space and time and the rule is that species must be complementary, improving the overall resource use efficiency. The key is to increase the system outputs per unit of resource input. Among outputs, it is important to consider not only the provisional ecosystem services (e.g., meat, milk), but also others including carbon sequestration, nutrient cycling, biological N fixation, habitat for wildlife and pollinators, and others. Complementarity in space indicates that plant species are exploring different soil layers to explore multiple pools of nutrients and water, have contrasting canopy architecture to intercept solar radiation, and have different tolerance to extreme weather, pests, and diseases. Spreading the risks makes the system more resilient. Complementarity in time indicates that plant species have different growth curves along the year, improving overall primary productivity. Therefore, improving species diversity is one way to improve adaptation to climate change. If the vegetation is heterogeneous, there is also opportunity to diversify the species of grazing (or browsing) livestock. On the mitigation side, diverse forage species in grazing systems may: improve forage productivity and quality with lesser fossil fuel inputs; decrease methane emission intensity; increase carbon sequestration; reduce nitrous oxide emissions and nitrate leaching. Diversification of forage species in grazing systems is critical in the sustainable intensification of livestock systems, and there are numerous examples of successful stories in temperate, subtropical, and tropical regions of the world. We must learn with the successful experiences and try to multiply when possible. The challenge ahead is to sustain a larger human population in a planet with limited resources and changing climate.

Session 66

Theatre 2

Carbon footprint of sheep farms in France: first results of the LIFE Green Sheep project

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Small ruminants farming generates greenhouse gas (GHG) emissions accounting for 6.5% of livestock emissions. For reducing their environmental impact, it is important to know and understand its level of emissions in order to identify areas for improvement. This is one of the objectives of the LIFE GREEN SHEEP project which aims at reducing the carbon footprint of sheep meat and sheep milk by 12% while ensuring farms' sustainability. For France, data from 670 sheep farms were collected as a first step of the project, representing various rearing systems. Data provide a good overview of the average sheep milk and sheep meat carbon footprint for France. Preliminary results indicate an overall GHG emissions from sheep meat and sheep milk of 3.0 kg CO₂e/kg FPCM (fat-protein corrected milk) and 42.0 kg CO₂e/kg carcass, respectively. These results differ between rearing systems. Nevertheless, the variability is mainly observed within each type of system. For instance, within dairy sheep systems from North Occitanie, the average GHG emissions of the 10% farms with the lowest emissions is 25% lower (1.8 kg CO₂e/kg FPCM) than the average emissions of the group (2.4 kg CO₂e/kg FPCM). This reflects the farms' differences in productive efficiency. Variations on carbon footprint are strongly related to herd management, consumption of inputs (concentrates, fertilizers) and days on grazing areas. The net carbon footprint has also been assessed to consider carbon storage. GHG emissions are offset by 54% and 26% for meat and dairy sheep systems respectively. These first results enable a better understanding of the correlation between carbon footprint and farm's practices and to identify areas of improvement for French farmers.

Greenhouse gas emission intensity of Irish Sheep meat: Irish Data Component of LIFE Green Sheep Project
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Sheep farming contributes to economic, environmental and social sustainability in rural areas where land use options are limited. It is important to determine the GHG intensity of sheep meat to identify and adopt practices that are environmentally sustainable, economically viable and socially acceptable. The main objective of the LIFE Green Sheep Project which involves 5 countries (Ireland, France, Italy, Romania, Spain) is to reduce GHG emissions of sheep meat production by 12% whilst ensuring farm level sustainability. Data were collected by the National Farm Survey which is part of the EU Farm Accountancy Data Network from 179 Irish sheep farms which were classified into extensive, semi-extensive, semi-intensive and intensive using the number of ewes per hectare. The mean emissions on these farms per kg of carcass weight (CW) and per sheep grazing area were 32.3 ± 1.6 (min 13.3 and max 193.5) kg CO₂ eq and $5,303 \pm 200$ (min 455 and max 16,777) kg CO₂ eq, respectively. The highest emission per kg of CW (54.2 ± 11.2 kg CO₂ eq) and the lowest emission per sheep grazing area (953 ± 225 kg CO₂ eq) were produced on extensive farms. The lowest emission per kg of CW (25.2 ± 2.1 kg CO₂ eq) and the highest emission per sheep grazing area were produced on intensive farms ($9,895 \pm 620$ kg CO₂ eq). The GHG emission intensity of sheep meat depends on flock management practices e.g. quantities of concentrate, inorganic fertilisers, lime, fossil fuel and electricity utilised. Including carbon storage in the assessment, mean GHG emissions intensity decreased to 21.6 ± 1.4 (min -59.1 and max 157.0) kg CO₂ eq/kg CW and $3,892 \pm 182$ (min -423 and max 14,989) kg CO₂ eq/ha. Using best management practices, sheep farming can potentially mitigate GHG emissions and increase economics returns.

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Theatre 4

Quantifying the impact of weather environment across three timescales on lamb performance

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Since current climatic impacts on livestock production are poorly understood and quantified, it is virtually impossible to predict the changes in production output likely to occur under various future climate change scenarios. This study aimed to use historical data to quantify the relationship between environmental weather conditions and lamb performance in a Dormer flock. Historical production and weather data collected on Elsenburg Research Farm in South Africa between 1972 and 2021 were used in the study. More than 9800 records from the Elsenburg Dormer stud flock for birthweight, weaning weight and lamb survival were used to calculate annual averages for each trait. Principal component analysis was used to condense seven weather parameters to meaningful principal components, each describing a set of environmental conditions. Factor loading scores for each of these components were then regressed on the lamb traits to determine which principal components (environmental conditions) significantly affected performance. Birthweight and lamb survival were significantly affected by weather conditions in the year preceding mating, with as much as 43.3% of the variation in birthweight being attributable to weather effects. Mating period weather only affected birthweight, but all three traits were influenced by weather conditions during lambing. Overall it can be seen that weather conditions affect lamb performance to varying degrees. These results clearly link predicted climate change impacts to a decline in sheep production efficiency.

Effect of sward type on urinary nitrogen excretion of late lactation dairy cows

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Nitrogen (N) concentrations in European waterbodies remain high, with agricultural sources contributing significantly. Recently, sward management factors have been demonstrated to be effective at mitigating N emissions. However, research on the effect of sward type on urinary nitrogen (UN) excretion under European conditions is limited. Therefore, the objective of this study was to investigate the effect of sward type on UN excretion of dairy cows. Nine rumen cannulated Holstein-Friesian cows were blocked based on pre-experimental milk production and body weight and randomly assigned to one of three treatments in a replicated 3×3 Latin square study design. The treatments were: perennial ryegrass (PRG) receiving 50 kg of inorganic N/ha/cut (GO); PRG-white clover (WC) receiving 25 kg of inorganic N/ha/cut (GC); and PRG-WC- plantain (PL) receiving 25 kg of inorganic N/ha/cut (GCP). The study consisted of three 29-day periods, each period consisting of 21 days of dietary adaption prior to an 8-day sample collection period. The cows were housed in individual stalls for the sample collection periods to facilitate the total collection of urine and faeces. Data were analysed in SAS using PROC MIXED. Cows offered GCP had greater urine volume when compared with GO and GC (76.8, 57.6 and 56.1 L/cow/day, respectively; $P < 0.001$). Cows offered GCP also had lower UN concentration when compared with GO and GC (3.5, 6.2, and 5.8 g N/L, respectively; $P < 0.001$). Finally, cows offered GCP excreted less UN when compared with GO and GC (255, 345 and 314 g UN/d, respectively; $P < 0.001$). The results of this study suggest that plantain inclusion in swards is an effective strategy to reduce UN excretion.

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Theatre 6

Dairy cows reduce drinking water intake when offered different fresh-cut pasture species

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Plantain (PL; *Plantago lanceolata*) can reduce nitrogen loss from pasture-based dairy systems. However, a reduced drinking water intake (DWI) has been observed with cows offered PL. A 2×2 cross-over design examined the effect of perennial ryegrass (PRG; *Lolium perenne*) and white clover (WC; *Trifolium repens*) mixture (PRGWC), or PRG, WC, and PL mixture (PLA) on DWI of lactating dairy cows. Feed and water intake were recorded over five days per period. Cow DWI was recorded hourly. Feed water intake (FWI) was calculated by adding the ad-libitum fresh weight intake of forage and concentrate intake (3 kg⁻¹ cow⁻¹ day⁻¹) and correcting for DM content. Total water intake (TWI) is the sum of DWI and FWI. Data were analysed using the MIXED procedure of SAS® Studio (edition 3.81). Forage DM content was lower for PLA compared to PRGWC (16.2 vs. 17.1 %, $P = 0.02$). Cows offered PLA had greater FWI, but lower DWI compared to PRGWC (106.3 vs. 93.1 L cow⁻¹ day⁻¹; 37.4 vs. 46.1 L cow⁻¹ day⁻¹, $P < 0.05$, respectively). However, no difference in DWI at any hour was found between treatments. Peaks in DWI occurred at milking and feeding time. No difference was found in TWI by cows between the PRGWC and PLA forages (139.2 vs. 143.7 L cow⁻¹ day⁻¹, $P = 0.25$). The TWI: forage DM ratio was greater for PLA compared to PRGWC (8.99 vs. 8.34, $P = 0.03$). A positive correlation existed with forage DM and DWI ($r = 0.75$; $P < 0.0001$). Lactating dairy cows offered a fresh-cut forage mix with a lower DM content have reduced DWI.

Factors influencing impacts of drought on grass yields in Dutch dairy farms

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In recent years Dutch dairy farms were confronted with several prolonged periods of drought, due to climate change. The precipitation shortages led to large reductions in grass yield, with impacts varying substantially between farms. The objective of this study was to identify farm characteristics and management practices that influenced effects of droughts on grass yields of Dutch dairy farms in the period 2016-2021. An online self-administered questionnaire with questions about farm characteristics and management practices was distributed among ±350 dairy farmers on sandy soils in the region Achterhoek in the Netherlands. From the 165 respondents, 113 farms were included in the analysis since these possessed complete farm records filled out in the Annual Nutrient Cycling Assessment (ANCA) model over the period 2016-2021. Grass yields used in the analysis were model estimates from the ANCA model. Daily precipitation records were collected for the same period from the weather station closest to each farm. First, the relative reduction in grass yield due to drought was estimated for each farm using a log-linear regression model for annual grass yield and farm-specific annual precipitation deficit. Second, farms were clustered based on main soil type. Last, farm management practices associated with the reduction in grass yield due to drought were selected in a succession of regression analyses. Results showed that grass yields were significantly lower in the years 2018, 2019 and 2020 compared to the years 2016, 2017 and 2021 ($P < 0.001$), with a 23% lower average grass yield in 2018-2020 compared to the other 3 years (8.9 vs. 11.6 t DM/ha, resp.). Impacts were largest on farms categorized as 'dry sand'. The predominant farm management practice influencing the relative reduction in grass yield due to drought was irrigation, besides other practices. It was concluded that droughts had a large impact on grass yields of dairy farms, and irrigation was the most important management factor alleviating negative effects. As water use for irrigation in dairy farms may be restricted in future, improvement of irrigation efficiency and alternative adaptation options besides irrigation should be further explored.

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Theatre 8

Generation of a livestock value of grasslands for the mitigation and adaptation to climate change impacts in Chilean Patagonia

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The objective of the work was to develop a livestock value (VG) index that allows assessing the plant communities present on farms, with which the carrying capacity and the carbon content of the soil can be determined more precisely. The study was carried out in one farm (2420 hectares) with extensive sheep production in the Magallanes region in Chilean Patagonia. The vegetal communities evaluated with the livestock value index and soil organic carbon (SOC) are Tussock grass, meadows, short grasses and Heath, which were determined through remote sensing processes with satellite images. For each class, exclusion cages of 0.18 m² were used to determine dry matter production and their quality (protein and energy). A cut was made at the end of the season to harvest the material produced, and the dry matter was analyzed. The livestock value formula applied was $VG = 1/101 (VP * VN + 100)$, where VP is the dry matter value and VN is the nutritional value of the vegetal communities. The results show that it was possible to differentiate between vegetal communities quantitatively by applying a livestock value to each paddock, giving a ranking of quality to compare with the rest of the paddocks. As a result of the differences found between communities the data for VG and SOC (%), 0 – 10 cm), respectively, were for Tussock grass (3.36 and 6.72%), meadows (20.33 and 29.10%), short grasses (3.30 and 5.85%) and Heath (1.90 and 13.62%). In this way, the VG obtained will be useful to determine the carrying capacity of a farm and also the carbon content of soil, depending on the respective vegetal community. The final product for farmers will be a map with all vegetal communities in farm and also a map with their VG or SOC.

The effect of sward botanical diversity on feed intake, growth performance and methane emissions of beef steers

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During ruminal feed digestion, methane, is released as a by-product of ruminal microbial fermentation accounting for a loss of dietary energy to the host, of between 2-12%, depending on the diet. There is growing interest in the potential of botanically diverse or 'multi-species' pastures to convey additional benefits including reduced fertiliser requirements, improved soil health and enhanced biodiversity, when compared with predominantly grass based swards. However, there is a dearth of information surrounding the impact of such botanically diverse swards on methane emissions in beef cattle. Therefore, the objective of this study, was to investigate the effect of two botanically diverse sward types, a 'multispecies' sward type containing six species i.e. grasses, legumes and herbs/forbs, vs. a perennial ryegrass/white clover sward on individual animal feed intake, animal performance and enteric methane emissions in Charolais cross steers (n=44). All animals were offered fresh zero-grazed forage daily, ad-libitum. Estimates of diurnal enteric methane and carbon dioxide emissions (C-Lock GreenFeed system) were collected together with daily feed intake throughout the duration of the 114 day experimental period. Individual animal feed intake was measured using the Calan gate system. Cattle were weighed fortnightly before feeding. Results of this study show there were no differences in individual animal feed intake (P= 0.2368) between treatments. Similarly, there were no differences in animal performance (P= 0.6134), or in enteric methane emissions between the two sward types studied (P=0.434). In conclusion, there is no additional benefit observed from the inclusion of herbs/forbs in a 'multispecies' sward type, when compared to perennial ryegrass/white clover in terms of animal intake, performance or methane emissions in beef steers. Data from this study will be the first to systematically compare the effect of botanically diverse swards on animal performance and enteric methane emissions in beef cattle.

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Theatre 10

Understanding impacts of scale changes on efficiency gains of pasture-based beef production systems in the Massif Central, France: a modeling approach

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Improving productive efficiency through better genetics, nutrition and management is often suggested as a solution to address sustainability challenges in livestock farming. In France, despite significant progress in animal breeding and an increase in concentrate use, the beef production sector slightly increased its carbon footprint and decreased its productivity of intermediate inputs (purchased goods and services) by 0.55% per year over the last 20 years. Therefore, the objective of this study was to understand why technical and environmental benefits achieved by improving efficiency at the animal scale do not necessarily transfer to higher scales. We compared feed efficiency (FE) of 2 beef suckler systems (with and without fattening phase) with Charolais cattle in the Massif Central at the animal scale (no reproductive animals) and the herd scale (with reproductive animals). These pasture-based systems are different in feed production, farm size and herd management practices. For each system, production of grass was simulated using the LINGRA model, whereas cattle production and herd management were simulated using the LiGAPS-Beef model. Preliminary results indicate the system without fattening had a FE of 159.2 g beef/kg DM at the animal scale and 31.0 g beef/kg DM at the herd scale. For the system with fattening, the FE was 81.5 g beef/kg DM at the animal scale and 40.0 g beef/kg DM at the herd scale. The system without fattening phase had a higher FE at the animal scale, because calves received a larger percentage of their nutrients requirements from milk. However, at the herd scale this system had a lower FE because calves were sold at lower weights. Subsequently, 2 more systems will be added to the comparison of efficiency (productivity, feed and nutrient use efficiency) and environmental impacts (GHG emissions, nutrient surplus) at the animal, herd and farm scale.

Assessing extensive pasture-based beef production in South Africa under future climate change

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Assessing the impact of climate change on beef production is crucial for developing customized adaptation and resilience measures. The objective of this study was to assess effects of climate change on extensive pasture-based beef production for Bos taurus, composite, Zebu indicine and Sanga cattle in 3 agro-ecological regions of South Africa under representative concentration pathways (RCP) 4.5 and 8.5. Genetic parameters of cattle were sourced from breed societies. Projected climate change data in the Green Book portal was used to generate future (2050) climate data. Future feed nutritional composition was obtained from literature. The results showed that baseline average daily gain (ADG) was higher ($P < 0.001$) (0.393kg/head/day) than the simulated RCP4.5 (0.188kg/head/day) and RCP8.5 (0.180kg/head/day) ADGs. Performance of Bos taurus and Zebu indicine declined more than other breeds while the Sanga was the most resilient, followed by the composite. Future climate change will have more adverse impact on cattle in Buffalo Berlin and Phalaborwa, while those in Bloemfontein will be least affected. Pasture quantity will limit cattle growth most in Phalaborwa, and pasture quality in Bloemfontein and Buffalo Berlin. These results can be used to adapt grazing strategies and pasture management and to breed for more resilient animals, which contributes to mitigating the expected decline in cattle performance.

Sheep grazing on peat grassland in a free-field photovoltaic system

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Renewable energies as well as the extensive management of peat grassland are a valuable contribution to climate change adaptation. Using the grassland of free-field photovoltaic systems for livestock grazing systems is an efficient way to maintain the swards and generate additional agronomic production. The present study was conducted in June and July 2023 at the 'Solarpark Lottorf', Schleswig-Holstein, Germany. We investigated the spatio-temporal patterns of ten GPS-collared sheep aged 3.5 ± 0.7 years (mean \pm SD) during active time and lying time in two evenly distributed sectors (with solar modules (SM) and without SM) within paddock. Additionally, we measured compressed sward height (CSH), the proportion of trampling and the distribution of faecal deposits. The paddock was divided into 80 5x5m gridcells for the analyses of CSH, the proportion of trampling and faecal deposits. GPS data for the analysis of the spatio-temporal patterns during lying time and active time were minute-wise retrieved from GPS collars. The sheep spent active time (associated with grazing) mainly in the sector without SM ($P = 0.0024$). Consequently, lying time as well as faecal deposits occurred mainly in the SM sector indicated also by significantly higher proportion of trampling ($P < 0.0001$). Pre-grazing CSH was higher in the SM sector while post-grazing CSH revealed no difference between sectors. SM can offer shade and protection to livestock. Combining renewable energies and pasture production systems appears to be a synergetic use case with a beneficial effect for animal welfare of grazing sheep.

Preliminary survey of the quality and pastoral value of different grazing areas in the park of the Campania Region

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The aim was a preliminary evaluation of pastures to calculate the animal load in two park areas of the Campania Region. The grazing areas locate in the Cilento national park (Monte Motola – SA) and in the regional park of the Picentini Mountains (Monte Polveraccio – AV). A survey of animal management and pastures was performed from summer to winter 2023. In the Cilento park the herds are kept free all year moving along the mountains with climatological trend. Pastures are located between 800 and 1700 m above sea level. High biodiversity was found; the trees are re-gaining ground, and the presence of animals keeps development the shrubs (*Rosa Canina* L. and *Rubus* L.) under control. This biodiversity affected on the pastoral value (VP). In the pasture at 700 m, 10 out of 14 plants detected had a pastoral index (IP) of 2 or 3 and VP of 43.4. Similar values were found in the pasture at 900 m where 7 out of 10 plants had an IP between 1 and 3 with a VP of 42.7. From the partial plant census, between 1000 and 1700 m we have found more uniform quality of pasture throughout of the year. As for the Monti Picentini park, herds are on pasture permanently as well but the animals are taken to the mountains at 1,400 m from July to November, the rest of the year they are kept at 500 m. Winter pastures quality is improved by overseeding and the analysis of the VP of two close fields with different exposure showed 10 species of which only 4 edible giving a VP of 39.5 and 14, 6 of them edible, with VP value of 36.3. The partial plant census at 1,400 m revealed a less constant pasture quality.

Greenhouse gas balance of livestock systems and carbon footprint of livestock products: two methodologies for assessing livestock contribution to climate change

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A wide range of methods to assess the impact of livestock activities on Climate Change (CC) at plot, farm, supply-chain, landscape, country or global levels does exist. The terms used in the literature can be confusing and are sometimes not used appropriately because a clear terminology is missing. We propose to distinguish two types of methodology on the basis of the most common used terminology: the “greenhouse gas (GHG) balance” and the “carbon (C) footprint”. This paper is based on a literature review of 235 scientific papers. “GHG and C balances” are based on an (eco)system approach. This methodology focuses on direct GHG emissions and C storage within the studied system boundaries. GHG balances focus on GHG emissions (CH₄, N₂O and CO₂) when C balances consider both GHG emissions and C storage in soil and trees. This methodology is generally applied at plot, farm or landscape levels and results are generally expressed per unit of surface area. The “C footprint”, also referred as the “emission intensity”, is based on a life-cycle assessment approach. This methodology considers both direct and indirect GHG emissions and C storage, it encompasses emissions associated with i) raising animals, including enteric fermentation, ii) upstream activities (feed, fertilizers, and other inputs production, processing and transport) and iii) downstream processes (manure and waste management, post-farm transport, processing and packaging of raw animal products). This methodology is applied at supply-chain level and results are generally expressed per unit of livestock product. Differences in goals, functional units, scopes, level of analysis, system boundaries, sources of GHG emission measurements or calculation (simple emission factors or complex mechanistic simulation models), taking account or not C storage and other modelling assumptions make it impossible to compare assessment results of the two methodologies.

Carbon footprint of Sardinian dairy goats reared in extensive farming systems

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The aim of this study was to estimate the carbon footprint (CFP) of 10 dairy goat farms representative of the traditional farming system of Sardinia, where the animals are reared in extensive grazing systems. Data were collected by interviewing the farmers, in a cradle to farm gate approach based on ISO14040:2006 and 14044:2006. The time boundary selected was a period of 12 months. Emission from enteric CH₄, CH₄ and N₂O from manure, CO₂ from energy and fuel consumptions, CO₂ from purchased and on-farm feeds emissions were quantified using the equations proposed by IPCC (2019). Greenhouse gases emissions (GHG) were expressed in CO₂ equivalent units assuming a 100-year time horizon (27.9 for CH₄, 273 for N₂O, and 1 for CO₂). The considered functional unit (UF) was one kilogram of fat and protein corrected milk (FPCM). On average, the farms have an area of 75 ha, ranging from 15 to 300 ha, a flock size of 175±89 head, and a milk production level of 234±176 kg of milk head⁻¹ year⁻¹. The assessment of the environmental impacts showed a mean total CFP value of 2.57±1.40 kg CO₂eq kg FPCM⁻¹ ranging from 1.12 to 5.90 kg CO₂eq kg FPCM⁻¹. The impact decreased increasing milk production level ($y = 87.103x - 0.684$; $R^2 = 0.8785$). Enteric CH₄ emissions were the largest source accounting for 65% of total GHG emissions, followed by emissions associated to purchased feeds (19%), manure management (13%), fuel and energy uses (2%), and on-farm feeds (1%). In summary, the results of this study show that the CFP values of goat milk from extensive farms are quite variable and decrease with increasing milk yield level and the area of land used by the farms. The authors acknowledge the RESTART and VERSOA projects for their financial support.

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Poster 16

Dry matter intake and calf performance on grass pastures in monoculture and silvopastoral systems in a subhumid tropical climate, Brazil

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Silvopastoral systems represent a promising alternative for the exploitation of land for the multiple ecosystem services that have the potential to provide livestock production. From the point of view of livestock farming, the insertion of leguminous trees in these productive arrangements can increase the efficiency of the use of natural resources, and can contribute to improving the nutritional value of the herbaceous component and animal performance. The objective was to evaluate forage consumption and animal performance of calves in different production systems. The experimental treatments consisted of a silvopastoral system (SSP) composed of *Urochloa decumbens* Stapf. with *Mimosa caesalpinifolia* Benth. in addition to grass monoculture, under a randomized block design, with three replications, during the evaluation period from August/2020 to October/2022. There was no significant effect ($p > 0.05$) of the type of pasture on animal performance, however, there was a higher ($P < 0.05$) forage intake in the SSP (3.29 kg DM kg BW⁻¹ d⁻¹), in relation to grass monoculture (2.54 kg DM kg PV⁻¹ d⁻¹). In the search for the intensification of livestock production in the Northeast region of Brazil, the exploration of the silvopastoral system with *Urochloa decumbens* and *Mimosa caesalpinifolia* may be an alternative, however, it is necessary to evaluate it over a longer period of time, aiming to verify whether this response pattern remains in order to obtain a safe management recommendation.

Carbon dynamics and greenhouse fluxes in subtropical grazing lands

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Grazing lands, including both rangeland and pastureland, cover about 10.7 million ha in Southeastern United States; have a significant impact on the carbon (C) balance in the region. Native rangelands in Florida extend over 3.1 million ha, and about 1.6 million ha of mostly non-forested areas cover from the center of the State to the Everglades, holding two-thirds of the State cattle. These rangelands provide many ecosystem services including food security, C sequestration, and climate change regulation. Florida's rangelands are grazed by livestock and often managed using prescribed fire or mechanical control of shrubs. This ecosystem is also subjected to a broad range of climatic variability, ranging from extreme droughts to extreme wet years where soils can be saturated with water for extended periods. Fire disturbs ecosystem functioning and C balance through direct release of C during ignition or indirectly by reducing ecosystem uptake capacity, altering soil physical-chemical properties and soil greenhouse emissions, and overall ecosystem warming potential. Droughts often reduce ecosystem C uptake; however, extreme rainfall events typical experienced during the summer months can cause rangelands to shift from a methane (CH₄) sink to a source. In addition, the presence of grazing animals also alters CH₄ emissions due to enteric fermentation. The objective of this study was to estimate ecosystem C balance and warming potential of representative subtropical grazing land ecosystems. We used a combination of eddy covariance and chamber measurements to estimate net ecosystem C exchange and greenhouse gas (CO₂, CH₄, and N₂O) balance. Mean pre-fire net ecosystem production (NEP) was -368 ± 41 g C m² yr⁻¹ and decreased to -181 g C m² yr⁻¹ during the year that fire was imposed. Despite the reductions in daily gross primary productivity (GPP) and NEP, the vegetation recovered its photosynthetic capacity 60 days following fire. Results indicated that subtropical grazing lands acted as strong C sinks sequestering ~ 3.2 Mg C ha⁻¹ yr⁻¹.

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Poster 18

Methane production in grazing sheep in the Magallanes region as a function of type of pasture

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The objective of this study was to evaluate the methane production (g CH₄) per day in grazing sheep as a function of the age of the animal and type of pasture, using different equations for the prediction of enteric methane emissions by sheep using an intercontinental database (Belanche et al, 2023; J Clean Prod). This research was carried out in Chilean Patagonia. Plant material was cut in three ecological zones to estimate the accumulated dry matter 2022-2023 season. Forage production (Ton DM/ha), chemical composition (DM, OM, CP, NDF, ADF, Lignin), Gross energy (GE, MJ/kg DM) and in vitro gas production technique were determined at 0, 3, 6, 9, 12, 24, 36, 48, 72 and 96 h, the DM disappeared (g/100g) was determined and methane (g CH₄/d) was calculated in adult ewes (50 kg), rams (104 kg) and young animals (34.7 kg live weight), using different prediction equations, using 6 equations Q1= $DMI \cdot 6.29 + 12.6 \cdot DMI$, Q2= $GHG \cdot (0.358 + 0.0299 \cdot GEI) + 0.0393 \cdot GEI$, Q3= $DMI + BW \cdot 2.47 + 10.2 \cdot DMI + 0.140 \cdot BW$, Q10= $I(PCC2006) \cdot 0.065 + GEI \cdot 0.05565$, Q11= $(IPCC2019) \cdot 0.067 \cdot GEI + 0.05565$, Q13= $DMI \cdot 7.82 + 12.7 \cdot DMI$. Equation Q2 presents the lowest concentration of g/CH₄/d, ($P < 0.001$) (being 18.33, 25.77 and 14.96 g/CH₄, for adult ewes, rams and young animals, respectively), equation Q11 presents the highest concentration of g/CH₄/d for adult ewes and rams (20.5 and 33.4 g CH₄/d, respectively), and is similar for young animals (14.5 g CH₄/d) with respect to equation Q2. Equation Q10 showed lower prediction performance, the IPCC_2019 equations (Q11) showed a higher gCH₄/d than Q1, Q2, and Q3 which was consistently associated with under-prediction. The Q3 equation $DMI + BW$ could be considered as the best applicable equation for predicting CH₄ production in young sheep, given that these variables are able to be monitored on farm, as a conclusion the appropriate universal equations (i.e Q1, Q2 or Q3) accurately predict CH₄ production across different forage diets composition without compromising prediction performance.

Pasture and animal production in integrated crop-livestock-forestry systems in the tropics

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In Brazil, there are several challenges regarding food production, which require rational and optimal land use, these problems are mostly associated with land and soil degradation, increasing costs of fertilizers, climatic factors, loss of biodiversity, pollution, lack of knowledge and poor management of the agricultural systems. One of the most innovative alternatives to overcome these problems and reach better productive indexes is the adoption of synergic and integrated crop-livestock-forestry systems (ICLF systems). Among the main benefits of the ICLF systems, we could list the diversification of income sources in the farming systems, intensification of rational land use, soil and water protection and conservation, nutrient cycling and soil fertility, N₂-fixing, reduction of the requirements for inorganic fertilizers, carbon sequestration and diminished greenhouse gas (GHG) emissions, increase biodiversity, habitat to wild animals and pollinators, environment protection, perception of the public, control and reduction of pests and diseases in plant and animals, reduction of the environmental impacts of the farming systems, animal welfare, ecological services, stimulus to preserve local genetic resources, among several other benefits.

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Theatre 1

Collaborations between research and industry in insect sector: case study illustrated by a black soldier fly producer

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All over the world, agriculture and agri-food industries inevitably generate losses. Bioconversion consists of recovering food-grade materials that are destined for destruction. Black soldier fly (BSF – *Hermetia illucens*) can valorize various food waste and can thus be produced all over the world in a circular economy logic. This solution responds to the growing need for sustainable ingredients and particularly proteins for animal nutrition (aquaculture, poultry, pigs, pets). Over the last decade, insect producers have invested on the development of zootechnical approach and industrialization in order to develop technically and economically efficient models. In parallel, several European researchers started investigating this new type of animal farming, especially through safety investigations, applicative fields and environmental evaluations, contributing to regulatory progresses too. Based on last decade experience, presenting general context and several practical examples (bilateral cooperations, H2020 European call, national projects, relationship through professional organizations) allows highlighting appropriate modalities, results, benefits and key-factors of success related to collaborations between Research and Industry. Insect products start being commercialized on the feed markets and many new farm projects are being under development. Although bioconversion by insects is an efficient, virtuous and natural-like process, some knowledge gaps remain uncompleted, such as reinforcing functional applications as well as environmental impact evaluation.

StayPrime: Prolonging Larval Shelf Life for Efficient Insect Farming Integration

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The rapid expansion of insect production faces significant hurdles in storage and transportation of live larvae, exacerbated by natural variations in egg output and short shelf lives of neonates, juveniles, and fully grown larvae. These challenges limit production capacity and drive-up operational costs, hampering market utilization. However, EntoPrime's groundbreaking solution, StayPrime, offers a game-changing approach, extending the shelf life of live larvae for weeks. In our trials, we successfully suspended Black Soldier Fly neonates, juveniles, and harvested larvae for 18, 15, and 7 days, respectively, under standard storage and transportation conditions. Remarkably, the weight of larvae remained consistent before and after suspension, indicating arrested larval development. Additionally, when fed with Chicken feed (CF), suspended neonates and juveniles exhibited higher growth rates compared to larvae fed directly on CF. Moreover, we observed a high survival rate among suspended neonates, juveniles, and post-harvest larvae. These findings underscore StayPrime's potential to address several critical needs in insect farming: i) StayPrime creates a buffer capacity for neonates, mitigating the impact of variations in egg output on production capacity. ii) It enables practical, cost-effective storage and transportation of juveniles under normal conditions, reducing operational inefficiencies. iii) It facilitates the seamless integration of live insects into traditional animal farms as high-quality premium live feed, opening up new market opportunities. In essence, StayPrime represents a paradigm shift in insect farming, offering unprecedented flexibility, efficiency, and quality in larval storage and transportation.

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Theatre 3

Automatization of abdominal color assessment in honey bees

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Abdominal coloration of honeybees was one of the first traits used to describe subspecies. The main disadvantage in using abdominal coloration as an indicator of the honeybee subspecies is strong environmental impact, and the subjectivity of coloration grading. Methods for objectively quantifying the abdominal pigmentation in honey bees have not yet been developed. We obtained four frames of honey bee brood from two colonies and incubated them at two different temperatures. One colony had workers exhibiting yellow marks on the abdomen, while the other not. We collected hatched workers and photographed abdomens. Images were analyzed using custom written R script to obtain vectors, that describes coloration on the length of abdomen. We used UMAP to reduce dimension of the vectors and developed classification procedure with the support vector machine method. UMAP did not distinguish individual abdomens according to experimental group. Trained classifier sufficiently separated abdomens incubated at different temperatures. We further improved the performance by preprocessing data with UMAP. However, we were not able to distinguish between experimental groups even though that the differences were seen by eye. We summarized vectors describing coloration in coloration index. We tested influence of brood origin and temperature on coloration index using ANOVA. Differences were confirmed between "yellow" and "gray" experimental groups. Our results indicate that environmental temperature in selected range during development does not seem to impact honey bee coloration significantly. The developed color recording protocol and statistical analysis provide a useful tool for quantifying abdominal coloration in honey bees with a great potential for differing between subspecies with distinct abdominal coloration.

Microbiome Dynamics and Waste Reduction Capabilities of Black Soldier Flies (*Hermetia illucens*) as Biosolid Consumers

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Biosolids, a by-product of wastewater treatment from households and industries, present significant disposal challenges in Australia, with annual production exceeding 300,000 tonnes and costs of A\$130 million. The Black Soldier Fly (BSF), *Hermetia illucens*, demonstrates effectiveness in reducing biosolid volume, potentially easing storage and transport issues and offering economic advantages through fertiliser valorisation. This study investigated the potential of BSF as efficient biosolid consumers, firstly by assessing the impact of diet composition and evaluating different life stages. We evaluated the waste conversion and assessed the impact of the BSF microbiome composition. Our results show that the substrate reduction efficiency reached 46%, with no significant differences observed among groups, diets, or colonies. We evaluated key parameters such as pH, temperature, number of larvae, total larvae mass, mean larval weights, mean larval feeding rate, and percent conversion efficiency, which reached a high of 23.9%. Microbiome analysis, conducted to investigate the microbiome dynamics of BSF utilizing 16S full-length sequencing, resulted in a distinctive separation between BSF gut microbiomes within different feeding groups and the biosolid diets. Our findings reveal the potential of BSF as efficient biosolid consumers and transformers, showcasing their ability to thrive across diverse diets while maintaining consistent substrate reduction. The observed microbiome variations shed light on the relationship between diet, life stage, and microbial community dynamics in BSF.

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Theatre 5

Exploitation of *Tenebrio molitor*, *Rhizopus oryzae* and *Trichoderma reesei* for the detoxification of contaminated bran with aflatoxins B1

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Abstract Recently, the *Aspergillus* toxigenic species, main responsible for the production of aflatoxins (AFB1) – a secondary highly toxic metabolite – have spread to temperate areas as a consequence of rapid climate change. The use of aflatoxin-contaminated crops is strictly regulated by legislation and contaminated crops cause major economic losses along the supply chain. The research aims to evaluate a combination of biological processes (insect rearing and fungi fermentation) to detoxify mycotoxin-contaminated crops and ensure food safety. Wheat bran was experimentally contaminated with increasing AFB1 levels (0-125-250-500µg/kg) and was used as a rearing substrate for *Tenebrio molitor* (Linnaeus, 1758) (yellow mealworm, YM) larvae for 44 days. At the end of the rearing period, residual insect farm waste (frass) was inoculated with *Rhizopus oryzae* (CCT7560) and *Trichoderma reesei* (QM9414) strains obtained from a culture collection furnished by the André Tosello Foundation, Campinas, SP, Brazil. The selected strains were selected for their known aflatoxin-degrading ability (Hackbart et al., 2014). Toxin levels did not significantly affect YM growth performance; moreover, the presence of AFB1 was detected, with a concentration of $4.10 \pm 0.9 \mu\text{g/kg}$, only in YM larvae reared on the highest level contaminated substrate. Frass collected at the end of the rearing phase resulted in a concentration up to $67.28 \pm 5.1 \mu\text{g/kg}$ of not metabolized AFB1. The subsequent fermentation process with *R. oryzae* and *T. reesei*, resulted in a complete depletion of AFB1 (not detected) in the frass. In conclusion, the high tolerance of YM to AFB1 and the apparent lack of bioaccumulation in YM body suggest the possibility to valorize AFB1 contaminated crops as YM rearing substrate while the further identified fermentation process allows to obtain a toxin-free soil conditioner for agriculture practices. Hackbart H.C.S., Machado A.R., Christ-Ribeiro A., Prietto L., Badiale-Furlong E. 2014. Mycotoxin Res 30:141–149 DOI 10.1007/s12550-014-0202-6

The effect of different wet fractionation methods on the yield of *Hermetia illucens* larval fractions and the distribution of the main constituents

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Hermetia illucens represent a potential nutritious, affordable food source. Although the larvae can be consumed in full biomass form, this does present some limitations. The larvae are rich in fat, which can limit intake. An alternative to consuming full biomass larvae is to fractionate it into the main constituents, i.e. fat, protein and chitin. Through fractionation it becomes possible to utilise individual constituents in food products. Insect fractionation has been described in a number of research papers, but there are still aspects that need investigation. The main purpose of this project was to evaluate different fractionation methods in terms of their effect on dry matter yield of different fractions and nitrogen and fat distribution. The larval biomass was fractionated using a combination of mechanical processing and one of eight additives. These were three organic acids, and an enzyme in combination with either water or one of two alkaline solutions. These additives were compared to water based control treatments at either acidic or alkaline pH. Each fractionation process produced four fractions: a protein rich solid, protein rich liquid, fat rich and chitin rich fraction. The dry matter yield of each fraction and the distribution of fat and nitrogen was determined. The data were subjected to an analysis of variance using Statistica version 14.0.1. The results indicated that fractionation methods had significant effects on fraction yields and constituent distribution. The treatment methods that were applied at an acidic pH showed increased fat recovery, whereas the alkaline pH treatments showed improved nitrogen recovery in the protein rich fractions. These fractionation methods could be employed in larval processing in the food industry and the choice of method would be based on the main larval constituent of interest. Further research is needed on the effect of fractionation methods on the structure of constituents and how that could affect their techno-functional properties and their potential application in food products.

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Theatre 7

Subcritical Solvents for the Biorefinery of Chitin from Black Soldier Fly Larvae (*Hermetia illucens*) – Expanding the Scope towards other Insect Biomass

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Chitin, which is present in the exoskeleton of insects, is interesting due to it being non-toxic, biodegradable, and biocompatible. Applications of this biopolymer include tissue engineering, drug delivery, medicine, biological control, and the production of renewable N-containing chemical building blocks. Traditionally, extracting and purifying chitin involves lengthy (72 hours) and harsh treatments with acidic, basic, and bleaching solutions. This results in large amounts of hazardous waste and degradation of the chitin quality. Advancing the current state-of-the-art, we recently have developed a sustainable biorefinery process with subcritical methanol to produce methyl esters, oligopeptides, and chitin from fresh black soldier fly (*Hermetia illucens*) larvae in 2 to 3 hours. To expand its applicability, we have investigated its potential to process other insect biomass, including holo- and hemimetabolous insect species and insect exuviae. Furthermore, alternative alcohols have been investigated as substitution for methanol. Our research demonstrates that this innovative method is effective in isolating chitin from all investigated types of insect biomass (e.g., crickets, mealworms: 75 – 77 % chitin in the purified product vs. ± 5 % in the starting material). At the same time, ethanol and butanol (chitin content > 65 %) have also been proven to be a viable replacement for methanol. Our findings thus represent a major advancement in the development of sustainable biorefinery technology for chitin isolation and purification from insect biomass.

Preliminary results on the control of Fusarium wilt on lettuce and tomato by insect frass

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Fusarium wilt is a common fungal disease caused by *Fusarium oxysporum* and poses a significant threat to various crops like tomato and lettuce. Control measures include using resistant cultivars, fungicides, physical methods, and practicing good soil management techniques. Insect frass is considered a soil amendment and alternative to conventional fertilizers and pesticides. The aim of this study was to assess Fusarium wilt suppression by insect frass on lettuce and tomato in greenhouse. Frass from *Hermetia illucens* larvae fed with Gainesville diet was produced at the experimental insect facility of the Department of Agricultural, Forest and Food Sciences. Part of it was heat treated at 70°C for 60 minutes. Both heat treated and untreated frass were blended into a peat substrate at 0, 1, 2, 5, 10, 20 % (v/v) dosages, then used to fill nursery pots in which tomato and lettuce seeds were sown. Plants were transplanted 4 weeks after in 2 L pots filled with a peat substrate previously inoculated with *F. oxysporum* f. sp. *lycopersici* or *F. oxysporum* f. sp. *lactucae*, causal agents of, respectively, tomato and lettuce Fusarium wilts. Five pots with one plant each were prepared for each blend, and placed in greenhouse. Disease development was assessed 4 times, after transplanting. Above ground biomass of plants was weighted at the end of trial. Data collected were analyzed according to Anova to identify which dosage of frass was effective. Untreated frass significantly reduced Fusarium wilts on both crops, while heat treated frass lost this suppression capacity, suggesting a potential role of native frass microflora in controlling the diseases. (Supported by Agritech National Research Center – PNRR CN 00000022).

Effects of insect frass fertilizer on tomato fruits untargeted metabolomics

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Insect (*Tenebrio molitor*, TM) frass and insect-based poultry manure has been tested as organic fertilizer in a field trial for the cultivation of tomato (Cv. Regina). The application of High-Performance Liquid Chromatography coupled with High-Resolution Mass Spectrometry (HPLC-HRMS) provides a powerful means to comprehensively assess the impact of various fertilization strategies on the metabolomic profile of tomato crops. Diverse fertilization protocols, including organic and inorganic sources, were implemented in tomato cultivation, and the resulting fruit samples were subjected to HPLC-HRMS analysis. Approximately 70 molecules were annotated, 30 in positive mode and 40 in negative mode and these compounds were used as potential indicators of the nutritional quality, flavour, and overall chemical diversity of tomatoes under varied fertilization regimes. The untargeted approach revealed significant variations in the metabolomic profiles among the different fertilization treatments. Distinctive changes in the levels of key compounds such as amino acids, organic acids, sugars, and phytochemicals were observed. The expression of these detected metabolites was increased by the fertilization treatments of chemical:poultry (50:50), chemical:poultry 75:25, chemical:frass 75:25, 100% organic, and 100% chemical. On the other hand, treatments such as 100% poultry, 100% frass, and chemical:frass 50:50 apparently reduce the annotated compounds' expression. This study underscores the importance of tailored fertilization practices in shaping the metabolomic landscape of tomatoes, offering practical implications for optimizing crop management strategies. The integrative approach of HPLC-HRMS in untargeted metabolomics facilitates an understanding of the complex interactions between fertilization and tomato crop biochemistry, contributing to the enhancement of crop quality, yield, and sustainability in agricultural practices. Supported by PRIMA2021 Advagromed project.

A Fly for a Fly – Frass to control cabbage root fly in cauliflower production systems in Belgium

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Particularly within European organic agriculture, there is a growing interest in using frass as an alternative fertilizer. The composition of frass varies based on factors such as insect species, breeding methods and processing techniques. Beyond its role as a fertilizer, frass possesses additional properties. Studies indicate that frass influences the rhizosphere of plants, exerting an impact on overall plant development. The cultivation of cabbage crops carries the inherent risk of infestation by the cabbage root fly (CRF). The larvae of this fly can inflict damage to the root system, leading to diminished crop yield or even complete plant loss. The phased-out use of plant protection products poses a challenge for both conventional and organic farmers, as they grapple with declining yields in their cabbage production systems attributed to the CRF. During the 2021 to 2024 growing seasons, Inagro and PCG conducted eight field trials on cauliflower, four in conventional and four in organic farming. The objective was to evaluate the potential of frass as an organic and cost-effective technique for controlling CRF. Controls included the use of entomopathogenic nematodes and an insecticide-based drench treatment before planting. The trials also explored various forms of frass, including pelleted frass from the black soldier fly (BSF). Throughout each trial, CRF density and plant losses were assessed weekly and yields determined at harvest. Results indicate that applying insect frass in cauliflower production enhances the plant's defense system. Specifically, the application of pelletized BSF frass at planting resulted in reduced damage caused by CRF larvae, leading to decreased yield loss compared to untreated plants. Over the eight field trials, the average cumulative plant loss due to CRF was 49% for untreated plants. In contrast, BSF frass application reduced this to 21%, and the chemical reference Spinosad resulted in a 19% plant loss. The application of insect frass in horticulture and arable crops shows great potential. Further research is planned to optimize application timing and explore its use in other crops

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Theatre 11

Assessment of insect frass as biofertilizer for fava bean cultivation under Mediterranean climate conditions

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The Mediterranean area is currently facing many agricultural challenges putting to a great extent food security into question. Alternative farming systems based on sustainable agricultural practices are highly necessary to adapt farming to the near-term climate change. The use of insect frass as a biofertilizer showed recently positive outcomes. Along these lines, our study aimed to evaluate the effect of frass of the black soldier fly (BSF), *Hermetia illucens*, on the fava bean cultivation in Morocco. Field trials were carried out in Ain Taoujdate according to a randomized block design. Based on the frass nitrogen (N) content and the N rate used as reference for fava bean crop, six treatments (with 3 replications each) were evaluated: negative and positive controls, 25, 50, 100 and 200%, corresponding to 1.09, 2.18, 4.35 and 8.7 tons of frass/ha, respectively; 100% representing the reference rate. Morphological, agronomic and physiological parameters were assessed, encompassing seed germination, plant development, yield and height, flowering timing, root and shoot dry matter, leaf count, leaf area and chlorophyll content. Our results highlight the effectiveness of BSF frass application for optimal fava bean growth under Moroccan climate conditions. Financial support for this research has been provided by PRIMA, a program supported by the European Union, under grant agreement No 2231, project CIPROMED (PRIMA Call 2022 Section 1 Agri-food IA).

Non-absorption of ferulic acid by the Black Soldier Fly larvae (*Hermetia illucens*)

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Black Soldier Fly larvae (BSFL) are recognized to be rich in macronutrients, especially proteins and lipids. Hence, BSFL have been more and more used for biowaste recycling to create value-added products. They especially make a suitable alternative source of protein for animal feed. BSFL also contains micronutrients such as vitamins and minerals. Recently it was demonstrated by our group that BSFL can bioaccumulate interesting concentrations of fat-soluble vitamins and carotenoids from the substrate they are reared on. This suggests that BSFL could bioaccumulate valuable concentrations of other phytochemicals. Among all the phytochemicals contained in plants, polyphenols hold a special place because numerous studies have suggested that they can have beneficial effects on health, whether in humans or in livestock. But no study has been dedicated to assess whether some polyphenols can significantly bioaccumulate in insects reared on polyphenol rich substrates. BSFL are commonly raised on wheat bran, rich in ferulic acid (FA) and caffeic acid (CA), compounds of nutritional interest due to their health effects, both on livestock and humans. The aim of this study was to determine if they can also bioaccumulate interesting quantities, from a nutritional point of view, of these phenolic acids. We therefore raised larvae on this substrate and measured the concentration of these compounds in larvae. We observed extremely low concentrations of FA in the larvae and we did not detect CA. Indeed only 1% of FA concentration observed in wheat bran was found in BSFL. the FA quantity measured in a whole larva was 5 times inferior to the theoretical quantity in just the BSFL gut filled with wheat bran. It is therefore clear that the larvae degraded almost all the FA present in the wheat bran. In conclusion, this study suggests that BSFL does not bioaccumulate FA and probably CA. On the contrary, it seems that the larvae catabolize it very efficiently, which suggests that they have a biochemical pathway dedicated to this elimination.

Agronomic potential of BSF frass in the cultivation of ryegrass using distinct soils

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This study aimed at evaluating the effects of frass derived from bovine slurry bioconversion by BSF larvae. Frass was applied in three soils (sandy, loamy and clayey textures) either alone or in combination with a mineral fertilizer and its effects on both soil and crop yield/quality (ryegrass *Lolium multiflorum*) were assessed in a pot experiment. The application of frass resulted in increased soil organic matter in all soils, with a more pronounced increase in the clayey (0.04 – 0.49%). A 3-fold higher enzymatic activity was observed with exclusive frass application in the sandy soil in relation to mixed or mineral applications. Both loamy and clayey soils rendered similar yields (around 16.8 g pot⁻¹). In the sandy soil, highest yields were verified in the presence of frass, alone or combined with mineral fertilization (around 11.9 g pot⁻¹), while exclusive mineral rendered 5.4 g pot⁻¹. It was concluded that frass can replace mineral fertilizers while maintaining similar yields. This work is funded by PRR, Mobilizing Agenda for Reindustrialization – InsectERA (C644917393-00000032).

Welfare Challenges in Small Ruminants

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Sheep and goats across Europe face unique health and welfare challenges according to their production and management systems. This review will offer an update on the state of small ruminant welfare research and current farm practices by touching on transport, disease and painful management procedures, Precision Livestock Farming (PLF) and sustainability. European regulations limit animal transport by road to eight hours before rest is mandatory. However, remote communities such as the Scottish Northern Isles send their livestock to the mainland via 10 to 14 hour long ferry journeys. These journeys have been studied to identify opportunities for welfare improvements such as drinker design and noise-reducing measures. Castrating male lambs is known to cause pain and to negatively affect lamb mental state. A study of a novel tool for castration, the ClipFitter, found reduced pain behaviour expression in young lambs (< 7 days old) to the same levels as uncastrated lambs. However, in older lambs (5-6 weeks old), the ClipFitter resulted in similar outcomes as the traditional rubber ring method. Gastrointestinal parasitism has been identified as one of the main welfare concerns for meat and dairy sheep in European stakeholder studies. Behavioural studies have found that lamb faecal egg counts of parasites have a positive association with standing inactive behaviour and are negatively associated with lying behaviour. This could indicate that parasitism increases discomfort behaviours, providing insight into the experience of parasitised sheep. PLF presents new opportunities to monitor ruminant health and welfare remotely and continuously. Studies have investigated the attitudes of European sheep and goat farmers towards PLF in countries such as Romania, Norway, and Greece. Bluetooth Low Energy has been validated for measuring the distance between a lamb wearing a beacon and a dam wearing a receiver. A study reported that this ewe-lamb distance is associated with ewe welfare indicators like lameness and poor fleece condition. Finally, animal welfare's relationships with carbon emissions are currently being investigated in small ruminants. Life Cycle Analysis (LCA) studies report that indicators of poor sheep welfare, such as the presence of mastitis or parasitism, can increase the carbon footprint of farms. Many new tools such as PLF and LCA are being applied to the assessment and improvement of small ruminant welfare. They may improve our understanding of sheep and goats' experiences on European farms and encourage interdisciplinary approaches to the improvement of their welfare.

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Theatre 2

Genomic analyses of parasite resistance traits in German crossbred Merino

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Gastrointestinal nematode (GIN) infections may compromise animal health, welfare and production in grazing sheep. To prevent too high GIN loads and parasitosis, anthelmintic treatment is applied. However, an increasing proportion of GIN has developed resistance against anthelmintics thereby decreasing treatment effectiveness in sheep. Breeding for an improved immunity of sheep against GIN is one sustainable and promising approach to combat GIN. Here we aimed at investigating the suitability of parasitological and immunological parameters as selection traits for breeding in German sheep. The experiment took place on three farms in Southern Germany in 2021 and 2022. Merino crossbred lambs (n=1129) were raised indoors and subsequently managed in two grazing systems (intensive/extensive). After four weeks of natural infection on pasture, blood and faecal samples were collected in late spring when the animals were on average 13 weeks old. Genotype data (50k) was available for all lambs. Parasite load (faecal egg count, FEC) of Trichostrongylidae spp. (FECt) and Nematodirus spp. (FECn) as well as immune globulin A concentrations in plasma (IgAp) and feces (IgAf) were quantified. Subsequent genomic analysis (multivariate mixed linear model) revealed significant heritability for all traits (0.36, 0.27, 0.27 and 0.52 for FECt, FECn, IgAf and IgAp, respectively). Low to moderate genetic correlations were found between the traits, however, larger datasets are required to draw inferences from these due to the high standard errors. The results imply that the traits are putatively suitable to select for improved immunity against GIN in German sheep but further research is required before deriving breeding strategies.

Health traits as a way to estimate faecal egg count in dairy goats? – preliminary results

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The aim of this project is to identify health and robustness traits in dairy goats, as there is currently no targeted recording of direct health traits in goats. Especially parasite load is of major concern, as dairy goats are often raised on pasture in Germany and treatment is limited due to resistances. This study evaluated the relationship of faecal egg count (FEC) compared with animal welfare indicators (AWI), body condition scores (BCS), linear body (LBM) and infrared thermographic measurements (ITM) in dairy goat herds in Germany. A total of 344 dairy goats were evaluated two to five times during two consecutive lactation periods. Mean FEC over all examinations was categorized in three groups according to their eggs per gram faeces (EPG): low (<500 EPG), medium (500<1499 EPG) and high (\geq 1500 EPG) FEC. Statistical analyses using SAS studio were used to calculate influencing effects for FEC. The high FEC group showed significant effects ($p<0.05$), having lower BCS, lower BW and lower ITM of the legs compared to the low FEC group. Further analyses are needed to define robust animals in terms of FEC. Additionally, further ITM's should follow, as this non-invasive measurement seems to be easy to implement on-farms and might serve as a direct health measurement in terms of FEC. The present research is funded by the German Federal Ministry of Food and Agriculture (funding code 28N-2-035-02).

Novel mRNA isoforms identified in abomasal tissue of sheep affected by *Teladorsagia circumcincta* gastrointestinal nematode

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Gastrointestinal nematode (GIN) infections lead to reduced productivity, economic losses, and welfare concerns in the global sheep industry. Transcriptomics using RNA-Sequencing (RNA-Seq) can effectively assess the differences in transcriptome profile and its impact on GIN resistance in sheep by identifying differentially expressed (DE) mRNA isoforms (DEI) generated due to alternative splicing. We aimed to assess the underlying genetics of GIN resistance in sheep by identifying DE novel and annotated mRNA isoforms using Large Gap Read Mapping of publicly available RNA-Seq data (NCBI GEO accession id: GSE63547). Abomasal lymph node tissue samples from Scottish Blackface lambs ($n=20$), classified as highly resistant (R; $n=10$) or susceptible (S; $n=10$) based on fecal egg counts, were analyzed using CLC Genomics Workbench ($|FC|>2$; $FDR<0.05$). Functional genomics analysis of DEIs was performed using Reactome ($FDR<0.05$). The analysis revealed 30 DEIs (10 up-, 20 downregulated in S), including 6 novel and 24 annotated DEIs between R and S sheep at 7 days post-infection (psi). At 14 days psi, 3 novel and 19 annotated (6 up-, 16 downregulated in S) DEIs were identified between R and S sheep. The WDFY4 involved in cell-associated antigens presentation ($|FC|=9.48$) and DOCK9 involved in GTPase-mediated signal transduction ($|FC|=2.05$) were the most significant DEIs upregulated in resistant sheep in the 7 and 14 d psi groups, respectively. Functional analysis using the list of DEIs revealed that 9 and 4 metabolic pathways were significantly enriched ($FDR<0.05$), and which are primarily responsible for cellular physiological events (i.e. glucose metabolism, apoptosis) in S sheep at 7 and 14 d psi, respectively. This study will provide a better understanding of the etiology of the disease by identifying key regulatory novel mRNA isoforms to enhance selection strategies for higher GIN-resistant sheep with high productivity.

Investigating farmers and crofters' experiences of unexplained lamb loss in the Highlands and Islands of Scotland

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Blackloss is the unexplained loss of lambs on extensive hill grazings in the Highlands and Islands of Scotland. High parasite burdens, predation, a photosensitisation disease known as plochteach, and poor nutrition are often given as presumed reasons for blackloss. Previously reported figures for annual lamb blackloss give an average of 18.6% loss between marking (June) and weaning (September). A questionnaire was developed to assess the experiences, impacts and understanding flock managers have of blackloss. Typology analysis was used to cluster respondents into three distinct groups: 1- very large extensive farms and Sheep Stock Clubs, 2- small-scale farms and crofts with access to common grazing areas, and 3- medium sized farms. The responses of these groups were subsequently analysed to see if their experiences and perceptions of blackloss differed with relation to lamb health challenges and predation impacts. The groups reported similar health challenges, apart from Group 1 which described a significantly higher proportion (1-20%) of their lambs suffer from plochteach than Group 2 (Kruskal-Wallis, $\chi^2(2) = 7.30$, $p = 0.02$). In terms of predators, Group 1 also perceived white-tailed eagles (*Haliaeetus albicilla*) as a much higher threat to their lambs than the other groups (Kruskal-Wallis, $\chi^2(2) = 7.47$, $p = 0.02$). It was observed that the groups shared similar attitudes, with many of the respondents considering that blackloss is inevitable and that predators pose a large threat to lambs, however the groups each agreed that reducing these losses is important and that understanding the causes would enable them to do so.

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Theatre 6

A longitudinal study into predisposing factors for lamb loss and photosensitisation on a Scottish hill sheep farm

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Blackloss is the unexplained loss of lambs on extensive Scottish hill grazings. Reported losses give an average of 18.6%, whilst anecdotal evidence suggests that plochteach (a hepatogenous photosensitisation disease) may be a contributing factor. The aim of this study was to investigate the role that plochteach has in blackloss. Data were available for 3,071 lambs born between 2014 and 2021 at Scotland's Rural College Auchtertyre Farm. Lamb presence/absence, together with liveweight of present lambs, were recorded at recording events using EID tags, and resulting count data was used to calculate blackloss. Plochteach was diagnosed through clinical signs of photosensitisation. The effect of lamb traits, plochteach, and dam and sire factors on blackloss were investigated. The eight-year average blackloss observed between tagging (May/June) to weaning (September) was 8.3%. During the eight-year period 5.1% of lambs were affected by plochteach. Prevalence of plochteach in the blackloss populations was 10.6%, indicating it as a cause of blackloss. Generalised Linear Mixed Modelling found that 6-week weight and plochteach significantly impacted blackloss ($p < 0.001$), with low weight lambs and those affected by plochteach at greater risk. Additionally, white skinned, single lambs raised in a hill environment were at a higher risk of photosensitisation compared with black skinned, twin lambs raised on improved grazing. These findings highlight the risk factors for plochteach and also the role both it and low 6-week weights play in lamb blackloss.

Functional Longevity of Cyprus Chios Sheep: Insights from Reproductive Data

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The functional longevity of Cyprus Chios sheep, a breed known for its prolificacy, adaptability and milk production, is crucial for sustainable sheep farming. This study investigates the breed's reproductive performance through key indicators such as litter size, litter deaths, milk yield, and risk of culling. Utilizing comprehensive reproductive data from a nucleus herd in the Athalassa farm in Nicosia-Cyprus, this study aims to provide insights into the factors influencing the longevity and productivity of Cyprus Chios sheep. The study analyzed reproductive records of 3216 ewes the Cyprus Chios breed over multiple lambing seasons dating from 1989 to 2022. Results indicate a significant variation in litter size born alive, with an average ranging between 1 and 3 lambs per ewe. Litter deaths were found to occur at a rate of 6.06%, primarily attributed to factors such as maternal care, genetic predispositions, and environmental stressors. Furthermore, the risk of culling among Cyprus Chios ewes was evaluated, considering factors such as age, reproductive performance, and milk production. Heritability estimates were 0.15 ± 0.01 for litter size live and 0.07 ± 0.01 for longevity, observed at the age the animal left the flock. In conclusion, functional longevity of Cyprus Chios sheep is influenced by various parameters including litter size, litter deaths, milk yield, and risk of culling. Understanding these factors is essential for implementing culling strategies, can enhance flock sustainability and profitability in sheep farming systems. Further research focusing on genetic selection and feeding management is underway to optimize the reproductive performance and longevity of Cyprus Chios sheep.

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Theatre 8

Assessment of Subclinical Mastitis in Cyprus Chios Sheep Using Milk Electrical Conductivity

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Subclinical mastitis (SCM) is a pervasive and economically significant condition affecting dairy sheep, including the local, high-yield Cyprus Chios breed. Early detection of SCM is crucial for effective management and prevention of the disease. This study aimed to evaluate the use of Milk Electrical Conductivity (MEC) as a parameter for the early detection of SCM in Cyprus Chios Sheep. A total of about 250 lactating Cyprus Chios ewes were included in the study, and milk samples were collected twice daily at monthly intervals throughout the lactation period (120 days). The MEC was measured using a specialized device, and a moving average was calculated to assess variations over time. Additionally, conventional methods, such as somatic cell count (SCC), were employed for comparison. Data were categorized into two groups based on the SCC range. SCC data below 400,000 cells/mL (Group 1) indicate healthier animals, while values above this threshold (Group 2) indicate a higher risk of udder infection. The results indicated a significant association (p -value = 0.0007) between the MEC values ($MEC_{Group1} = 8.12 \pm 0.04$ and $MEC_{Group2} = 8.50 \pm 0.10$) and the 2 SCC groups. In addition, the Pearson's correlation coefficient (r) between EC and SCC in Group 2 is approximately 0.15 indicating a positive linear relationship (p -value = 0.02). Additional data could potentially enhance the robustness and reliability of the correlation analysis. Therefore, further data collection efforts will be pursued to better understand the relationship between EC, SCC, and SCM in Cyprus Chios Sheep. This non-invasive and real-time monitoring approach could provide valuable insights for implementing timely intervention strategies and improving overall udder health in Cyprus Chios Sheep herds. In conclusion, the utilization of MEC proves to be a promising and efficient method for the early detection of SCM in Cyprus Chios Sheep. Additional research and validation are needed to fully ascertain the practical utility of MEC as a tool for improving udder health management in dairy sheep populations.

Metagenomic and Genome-Wide Association Analysis in Ewe Fertility: A Comprehensive Study

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Artificial insemination success rate in sheep is low. Recent studies focused on the role of vaginal microbiota in ovine fertility, previously noted for its importance in human fertility and its association with reproductive health in cattle. This research used nanopore sequencing to assess the ovine vaginal metagenome impact on fertility and associated factors. Our objectives were: 1) Characterizing the microbiota and gene composition (COG & KEGG); 2) Identifying herd and breed influences on microbial profiles; and 3) Assessing the correlations between these microbial-genetic patterns and pregnancy outcome. The study involved 297 ewes across four herds from three Spanish breeds. Pre-insemination, vaginal exudates were collected and processed for microbial DNA sequencing using nanopore and adaptive sampling technology. Analysis included basecalling, barcode filtering, host DNA removal, and taxa assignment with SqueezeMeta pipeline. Microbial diversity was assessed using composition analysis at the genus level, COG, and KEGG, beta diversity (β -div) through PCA and PERMANOVA, and differential abundance analysis (DAA) to explore associations with pregnancy, adjusted by herd differences. Centred-Log-Ratio and Size-factor transformations were used for β -div and DAA, respectively. Microbial composition showed a general trend of predominant taxa such as *Escherichia*, *Staphylococcus*, and *Shigella*. COG and KEGG assignments revealed genes and pathways associated with microbial-host interactions and immune responses. β -div revealed the significant impact of herd and breed on microbial composition. DAA revealed *Fusobacterium* and *Campylobacter* to be more abundant in non-pregnant ewes. Conversely, genes like COG4678, K01185, and K14744 were enriched in pregnant ewes, suggesting a protective role in pregnancy. These findings highlight the intricate relationship between microbiota and reproductive health, with implications for herd management practices.

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Theatre 10

Differences in the mammary gland transcriptomes between sheep with different SRLV infection status and TMEM154 E35K genotype

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Small ruminant lentiviruses (SRLV) infect animals worldwide. Differences in genetic susceptibility of sheep to SRLV infection are evident by the association of breed and genetic markers, especially an amino acid substitution in the transmembrane protein 154 (TMEM154 E35K), with SRLV infection status. Sheep homozygous for the K allele (KK) are less susceptible than sheep with one or two E alleles (EK or EE). However, the effect of TMEM154 E35K seems to depend on sheep breed and/or SRLV genotype, and it is very likely that additional genes/factors play a role in the SRLV susceptibility of sheep. The study aimed to investigate differences in the mammary gland transcriptomes between sheep with different serological SRLV infection status and TMEM154 E35K genotype. Merinoland ewes (> 3 years old) were serologically tested for SRLV infection and genotyped for TMEM154 E35K. Three groups were identified: 1/SRLV positive, TMEM154 EK (posEK); 2/SRLV positive, TMEM154 KK (posKK); 3/SRLV negative, TMEM154 KK (negKK). Mammary gland samples for RNAseq were collected post-mortem from 9 ewes. Differential expression analysis was conducted using DESeq2 v.1.42.0. Comparison of sheep negKK and posEK revealed enriched GO terms and KEGG pathways, including those related to MHC class II protein binding, ECM-receptor interaction, human papillomavirus infection, focal adhesion, PI3K-Akt signaling pathway. DEGs between posKK and posEK were enriched in different GO terms including those related to tolerance induction, microtubule bundle formation, cilium assembly and organization, cell projection assembly, microtubule-based process, and tube and epithelium development. Transcriptomic results may help explain differences in the genetic susceptibility of sheep to SRLV infection. This research was supported by the National Science Centre, Poland (grant no. UMO-2020/39/1/NZ9/01304).

Identification of recessive lethal mutations in sheep using homozygosity deficiency

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Livestock, similar to humans, harbor deleterious mutations within their genomes. These recessive mutations, when present in the homozygous state, can lead to fetal or neonatal lethality, as well as morphological defects. This reduces the reproductive success of female breeding animals and negatively impacts health and welfare. Inspired by reverse genetics approaches in cattle, we investigated the genomes of two dairy sheep populations, searching for haplotypes that are rarely or never found in the homozygous state. These deficits in homozygotes potentially indicate the presence of recessive lethal mutations. Utilizing 50k-SNP genotyping data and pedigree information, we previously identified 13 independent haplotypes exhibiting a deficiency in homozygotes. Through whole-genome sequencing and targeted matings to generate homozygous animals, we uncovered three recessive loss-of-function mutations responsible for neonatal or juvenile lamb lethality. A nonsense mutation in the *CCDC65* gene disrupts ciliary function, leading to respiratory failure and lamb mortality prior to weaning. Another nonsense mutation in the *MMUT* gene disrupts methylmalonic acid metabolism, causing lamb death within the first five days of life. Finally, a single base pair duplication in the *SLC33A1* gene results in fetal losses and neonatal mortality. This study demonstrates the effectiveness of reverse genetics in identifying genetic defects in sheep. Implementing specific management strategies for these haplotypes/variants within dairy sheep breeding programs has the potential to significantly improve overall fertility and lamb survival rates.

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Poster 12

Problems related to ear-tagging goats

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It is common practice, and in many countries mandatory, to ear-tag goats. This practice is often believed to be benign, but scientific evidence is scant. Hence the objectives of this study to determine the prevalence of ear-tag related problems among goats kept by hobbyist and farmers, and to survey their opinion on different identification methods. The online survey was disseminated by convenience sampling among keepers of hobby goats in Flanders and the Netherlands. This questioned ear-tag related problems, opinions on various aspects of ear-tagging and interest in alternative identification methods. In addition, ear-tag related problems were scored on 3 organic and 5 conventional goat farms. These farmers were surveyed too. The survey was filled out by 71 hobbyists, providing information on 380 goats ear-tagged for >3 months. Twenty-five (35.2%) respondents refuse to ear-tag certain goats, and 37 (52%) respondents noted clear behavioral changes after ear-tagging. The prevalence of ear inflammations depended on type of tag: 39% for buttons, 24% for plates, and 22% for stripes. The desired situation (intact and readable ear-tag not associated with visible ear lesions) was present for 77% of the strips, 58% of the plates, and 53% of the buttons. Respectively 58.1% and 68.6% of the respondents gave the lowest satisfaction score (1/10) for the ear-tags in general and for animal welfare specifically. Most respondents (84.9%) stated to be willing to pay a premium price for marking their goats with a microchip instead of an ear-tag. Respectively 601 and 3040 ears were assessed on organic and conventional farms. The desired situation was observed for 81% (organic) and 72% (conventional) of the ears. Chewed ear-tags were not seen in organic farms, but were common (11%) in conventional farms. Inflammations were more common on organic (8%) than conventional (5%) farms. Satisfaction scores were higher among farmers than hobbyist. Animal friendlier means of identification are needed, in particular for goats kept by non-professionals

A pilot study of resistome characterization by using long read whole genome sequencing: from sheep faeces to soil fertilised with composted manure

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In sheep farming, the presence of antibiotic resistance genes (ARGs) in animal manure, and the composted manure may threaten environmental safety. Herein, we aim to better understand and characterize the resistome profiles in microbial communities found in two lamb feedlots. Total DNA extraction was performed using Quick-DNA™ HMW MagBead Kit (Zymo Research) from samples of pooled animal faeces (n=10), manure (n=4), and composted manure (n=4). Furthermore, samples from soil before (n=5) and after compost application (n=5) from one of the feedlots were also analysed. Metagenome sequencing was performed using a MinION device (Oxford Nanopore), with v14 chemistry and R10.4.1 flow cells, pooling previous DNA samples into one sample for each feedlot: animal faeces (n=3; an additional sample with sick lambs was also analysed), manure (n=2), composted manure (n=2), soil before and after compost application (n=2). We used the EPI2ME platform and ARMA (Antibiotic Resistance Mapping Application) pipeline to obtain gene annotation based on CARD database. Statistical analysis were performed using ResistoXplorer online software. Abundance in equivalent genome copies of ARGs per Gb of reads in a sample was calculated. A higher prevalence of tetracycline resistance genes (over 50% of the total ARGs), followed by drug pump efflux genes (around 30%) were found in faeces. In manure and soil, drug pump efflux genes were found predominant. Differential abundance analysis showed significant differences between feedlots at manure and composted manure level. However, no differences were found between samples from soil before and after compost application. Long reads enabled the detection of ARGs abundance in mobile elements such as plasmids or MGEs.

Prevalence of gastrointestinal parasitic infections in goat flocks in the Coastal Valley of Lima, Peru

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This study aimed to assess the prevalence of gastrointestinal parasites (GIP) and parasite burden in goats grazing stubbles from Lima, Peru. A total of 259 goat fecal samples were collected from 10 herds in two districts of Lima (Huaral and Aucallama) during winter 2023. The presence of GIP eggs/oocysts was determined using the sedimentation-flotation method and modified McMaster technique for counting. Descriptive statistics were used to establish parasite prevalence, and the Chi-square test was used to examine the association between prevalence and risk variables such as sex, and age. The data were processed using R software v. 4.3.1. The overall prevalence of GIP in the present study was 99.2 %; with prevalence of 98.8%, 44.4%, 34.6%, 6.2%, 1.9%, and 1.2% for *Eimeria* spp., Strongyle-type eggs (STE), *Skrjabinema* spp., *Moniezia* sp., *Trichuris* sp., and *Fasciola hepatica*, respectively. Maximum eggs/oocysts per gram (Epg/Opg) of *Eimeria* spp., STE; *Skrjabinema* spp. and *Trichuris* sp. were 26500, 16500, 400, and 50, respectively. Sex and age had a significant association ($p < 0.05$) with the presence of STE, where females over four years old had more prevalence. Likewise, there was an association ($p < 0.05$), between age and the presence of *Skrjabinema* spp where goats over four years old had more prevalence. *E. alijeivi*, *E. apsheronica*, *E. arloingi* and *E. caprina*, were the most frequent coccidia parasites. It was concluded that in the goat flocks of the Coastal Valley of Lima, the prevalence and parasite burden of *Eimeria* spp. was the highest, suggesting the need to implement parasite strategic control and prevention programs.

Assessment of ovine reproduction performances under solar grazing in Belgium: a pilot study

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The objectives were to assess the wellbeing and reproduction performances of ewes grazing under solar panels. Two groups of 20 ewes of similar age, weight and body score condition were included in the study. Rams were tested for semen quality before being used for reproduction. Ewes and rams were allocated to either solar grazing or conventional grazing. Wellbeing (flock behavior, comfort and locomotion) was evaluated 3 times a week. Mating was assessed via mating mark and harness on rams. The reproduction period lasted for 35 days. Then rams were removed from pastures, and 35 days later, ultrasonography was performed to detect pregnancy, to count foetuses and to estimate gestational age. Over the study period, all animals did not show alteration of wellbeing. Data analysis showed similar trends in marking of ewes in both grazing conditions. All ewes were mated during the first 15 days. Some went mated again afterwards : 6 in the control group; and 4 in the solar grazing group. Ultrasonography revealed 16 pregnancies in the control group, and 20 in the solar grazing group, i.e. a fertility of 80% and 100%, respectively. Number of foetuses per pregnant ewe were 1.9 +/-0.5 in the control group and 2.1 +/- 0.7 in the solar grazing group. Expected productivity is 30 lambs for the 20-ewes control group, and 42 lambs for the 20-ewes solar grazing group. Combining agriculture (crop or animal production) and renewable energy is an innovative way to use lands. However, the impact of solar panels on animals or vegetable production must be documented to better advise field actors.

Effect of progressive weaning on growth performance and cortisol levels of D'man lambs

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This study aimed to explore the effect of progressive weaning (PW) on D'man lambs' growth and cortisol levels in intensive farming conditions. A group of 8 males and 8 females (3 months, 14.9 ± 2.4 kg) underwent a two-stage weaning process. During the first stage, lambs were separated from ewes during the day and reunited at night. In the second stage, they were completely separated. Weight measurements and blood samples were taken before, during, and after weaning for cortisol analysis. Results revealed that lambs' growth speed declined temporarily (P<0.0001) during the first stage of PW, with an average daily gain (ADG) dropping from 121±27 g/day to -70±26 g/day. It then rapidly increased (P<0.0001) to an ADG of 191±26 g/day from 25 to 32 days post-weaning leading to a compensatory growth to reward the decrease of ADG resulting in a significant increase in lamb weight during this period (P<0.01). Cortisol levels showed no significant variations during both stages and after weaning (P>0.05). However, a higher cortisol value (P<0.05) was exhibited before weaning which could be attributed to age differences during the experiment. During weaning 4 out of 16 lambs exhibited diarrhoea, considered as a sign of stress-induced sickness. Cortisol, weight, and growth fluctuations suggest that PW may have temporarily impaired lambs' welfare.

Use of whole-genome resequencing for the study of genes associated with small ruminant lentiviruses susceptibility in two Spanish dairy sheep breeds

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Small ruminant lentiviruses cause a multisystemic inflammatory disease in sheep called maedi-visna (MV), resulting in high economic losses for the sheep dairy industry. Spain shows a high individual prevalence of MV (~50%) but with significant differences between the breeds analyzed. This study aimed to evaluate the genetic diversity of three candidate genes associated with MV susceptibility in two dairy breeds reared in Spain, Churra and Assaf. From whole-genome resequencing datasets (47 Churra vs 18 Assaf), a total of 677 variants were identified across three candidate genes: TMEM154 (638), CCR5 (4), and TLR9 (35). Among the genetic variants detected, 21 were predicted to produce a high or moderate impact on the corresponding transcriptional unit. For the CCR5 and TLR9 genes, we identified 3 and 4 missense variants, respectively. None of them showed significant differences between breeds. For TMEM154, we found 6 variants with relevant functional consequences (1 frameshift, and 5 missenses). Using a Cochran-Armitage trend model, significant differences were observed for the TMEM154 rs408593969 SNP between Churra and Assaf ($P=2.62E-06$). This mutation causes an amino acid substitution (K/E) at position 35 of the TMEM154 protein associated with the serological MV status in USA sheep populations. In particular, the KK proteotype, found in a higher frequency in Churra, has been related to a lower MV susceptibility and may be associated with the lowest prevalence of MV in this breed. These preliminary findings highlight the potential importance of rs408593969 and the need to further investigate its role in MV susceptibility mechanisms in the breeds analyzed.

Influence of a prepubertal dietary protein restriction on ewes' blood transcriptome in response to an intramammary inflammatory challenge

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This study investigates the impact of a prepubertal dietary protein restriction on the blood transcriptomic response to an intramammary inflammatory challenge in dairy ewes. For this purpose, forty Assaf ewe lambs aged between three to five months were divided into two groups: Control (C) and Nutritional Protein Restriction (NPR), with NPR animals receiving a diet lacking soybean meal supplement. Following this challenge, all animals were kept together and fed the same diet. At the end of their first lactation, 24 ewes (11 C; 13 NPR) were subjected to an intramammary gland infusion with *E. coli* LPS. Transcriptomic analysis performed on RNA-Seq datasets resulting from blood samples collected 24 h after the LPS challenge identified 87 differentially expressed genes (DEGs) between the C and NPR groups. Functional enrichment analysis highlighted that DEGs overexpressed in the C group were associated with molecular functions related to phospholipid phosphatidylinositol (PI) compounds, known for their immune suppressor properties in murine models. On the other hand, the DE genes with higher expression in the NPR group were related to cholesterol biosynthesis, with the SREBF1 gene being a key regulator of these pathways. SREBP-1a has been found to link lipid metabolism to the innate immune response. This preliminary analysis suggests that dietary restriction during development may influence the inflammatory response in dairy ewes, highlighting the potential biological mechanisms involved. Further research should be undertaken to fully understand the mechanisms underlying the results described here.

Correlation between genetic values for resistance to gastrointestinal nematode infection and growth traits in sheep

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Genetically improving sheep resistance to gastrointestinal nematodes (GIN) is an effective sustainable approach. However, it is important to understand the relationship between resistance to GINs and traits evaluated in breeding programs. In this study, we estimated correlations between Expected Progeny Differences (EPDs) for resistance to GIN and growth traits from the Canadian breeding program (GenOvis) for a commercial farm that has been selecting for resistance to GIN for the past ten years. The EPDs of 14,739 animals of Rideau Arcott sheep breed from the growth traits evaluation system (lamb survival (LS), birth weight (BW), 50-day weight (50W), gain 50–100 days (G100), ultrasound fat (UF), ultrasound loin area (UL)) were used. EPDs for fecal egg counts (FEC) measured using the Triple chamber (FECTC) and McMaster (FECMC) methods for the same animals were estimated using a two-trait repeatability animal model in ASREML software. The significant low negative correlations between LS, 50W, G100, and UF and FECTC (-0.14, -0.03, -0.12, and -0.12, respectively, SE=0.01) and FECMC (-0.15, -0.04, -0.13, and -0.11, respectively, SE=0.01), indicate that the selection to reduce FEC may indirectly lead to a slow increase the other traits. The significant low correlations between BW and FECTC (0.08 ± 0.01) and FECMC (0.09 ± 0.01) indicate that reducing FEC may slowly reduce BW. The correlations between UL and FEC traits were not significant. As all correlations were low, selecting for resistance to GIN is not expected cause any substantial changes in other traits currently selected for in Canada. Next, genetic correlations will be estimated to confirm these preliminary findings.

Session 68

Poster 20

Effect of shearing ewes during the first third of gestation on body weight and testicular development of male offspring

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Shearing ewes in early gestation improves lamb's weight at birth and weaning and survival. However, the long-term effects of early shearing on male progeny have not been studied. Shearing ewes in the first third of gestation (PS) and control unshorn ewes (U) were evaluated for testicular development (scrotal circumference (SC), testicular weight (TW) and testicular volume (TV)) and body weight (BW) of their lambs, whether singleton (S) or twins (T), from weaning to 18 mo-old. Seventy-nine male lambs reared on pasture were divided into four subgroups, single (SPS and SU) or twin (TPS and TU) born to PS or U-ewes, respectively. Considering date of birth, 24 lambs (SPS, n=6; SU, n=6; TPS, n=7 and TU, n=5) were castrated in early spring (non-breeding season, N-BS), with an equal number undergoing castration in early autumn (breeding season, BS). The TW and TV were measured, in N-BS (12.7 mo-old) and BS (18.2 mo-old). The BW and SC were recorded every two weeks, from weaning to the end of the experiment (3.5 to 18 mo-old). Data on SC, TW, TV and BW (repeated measures analysis) were analysed with PROC MIXED from SAS including shearing time, litter size, and interactions as fixed effects. The TW, TV and SC did not differ among treatments at either evaluation moment (N-BS and BS). The BW of lambs born to PS ewes was higher ($P=0.02$) than the U-group (34.6 ± 0.7 vs 32.2 ± 0.7 kg; mean of the whole experimental period). Shearing in the first third of gestation increased single or twin lamb's BW throughout the experimental period, however, testicular parameters were not affected.

Identification of infectious abortion issues and the changes in flock performance before and after diagnosis
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Abortions caused by infective agents such as *Toxoplasma gondii* and *Chlamydia abortus* can cause significant issues for sheep through reduced pregnancy rates, increased veterinary bills and the associated additional economic costs. Initial diagnosis time in flocks varies depending on management and the farming system but for many flocks the diagnosis of an infectious abortion agent only comes after a significant abortion event. This study aimed to track the changes in lambing percentages before, during and after an abortion diagnosis to understand the effect of abortion outbreaks on flock performance. The 160-ewe flock where the data was collected commenced lambing indoors from March 10th with a target of >95% of mature ewes (ME; >18 months of age) mated lambing and >80% yearling ewes (YE; <18 months of age) mated lambing. Infective abortive agents were identified following diagnosis in an Irish Department of Agriculture and Marine veterinary laboratory using aborted foetus and placenta samples. During the year of diagnosis, 86% of ME mated lambed at full term while 62% of YE mated lambed at full term. The pregnancy rate for the flock had been dropping from the previous lambing and was 91% for ME in the lambing immediately pre-diagnosis lambing and was still below target at 93% two years after diagnosis. Four years after the diagnosis and implementation of a management plan involving vaccination and antibiotic treatments the number lambed at full term exceeded the targets for the first time when 98% of ME and 85% of YE ewes lambed at full term. This case study clearly demonstrates that long-term effects of abortion issues due to infective agents even after the introduction of a treatment strategy. These effects can be identified at low levels initially where monitoring of pregnancy rates takes place and any drop in pregnancy rates should be investigated to reduce the long term effects of flock health issues.

Session 69

Theatre 1

In utero heat stress alters calf phenotype: the role of programming

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Exposure to heat stress in late gestation results in a phenotype of low productivity, reduced immune status and poorer reproductive performance in the dairy cow for the next lactation, and is an example of reversible programming. However, the impact of maternal heat stress late in fetal development has profound impacts on the calf, including arrested mammary gland development, slowed ovarian growth and lowered follicular reserve with substantial implications on productive potential and survival (i.e., milk production and longevity). Because these phenotypic effects are passed on to the offspring of the animals originally heat stressed, it is likely that epigenetic processes, such as methylation, drive the differences observed. Indeed, these and other tissue level effects are associated with shifts in methylation patterns in coding and non-coding regions of multiple genes that are consistent with altered expression. For example, in utero heat stress results in differential methylation of pathways associated with transcription, immune function, cell signaling, enzyme activity, cell cycle, and development among others in liver tissue of newborn bull calves. In the mammary gland of first lactation cows that had been heat stressed in utero, pathways related to protein binding, phosphorylation, enzyme and cell activation, and cell signaling were differentially methylated, and over 50 common genes were affected in both liver and mammary gland despite tissue, gender and age differences. While connections between methylation status and gene expression are limited, the pathways affected are comparable. Additional phenotypic observational evidence of transgenerational effects of environmental factors, especially heat stress, can be found in studies of season of birth effects on productivity, reproductive performance, and longevity. Collectively, controlled and observational data indicates that late gestation heat stress programs a low productivity phenotype and that through epigenetic mechanisms, the low productivity phenotypic changes persist in subsequent generations.

Transgenerational effects of heat stress in dairy sheep

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Previous studies on dairy cattle showed that the effect of cows' birth month, detected on the breeding values of granddaughters, is likely due to heat stress (HS) suffered during the pregnancy. In the framework of SCALA-MEDI project, the present study aims to investigate the transgenerational effect of HS in dairy sheep. In Sarda breed, births of ewes are concentrated in November and December. However, a differential exposure to HS, along gestation (summer-autumn) and across years or locations, can be observed. Pregnancies, lactation and climate data were recorded from 1980 to 2022 in 2 experimental farms located in the North and in the South of Sardinia. Milk yields of 4,931 genotyped ewes born from 1999 to 2022 were used to estimate GEBV by an animal model based on genomic relationships. GEBV of lactating ewes were analyzed with a linear model that included, as fixed covariates, the Temperature-Humidity Indexes (THI) recorded during pregnancies of dam (D), granddam (GD) and great-granddam (GGD) and GEBV of sires to adjust for non-maternal genetic effects. As a measure of HS, the average THI and the count of days where THI>23, were considered in the 1st, the 2nd and the 3rd thirds of the gestation. Both the HS measures led to the same results. No significant effects were detected in the 1st third of gestation. A significant negative effect of HS was observed in the 2nd third of GD pregnancy only. In the last third, significant negative effects of HS were estimated for D, GD and GGD. The most negative effect was observed for GGD and corresponded to -0.31 liter (-0.013 s.d.u.) per unit of average THI. These results suggest that epigenetic modifications in germ cells of embryo due to HS can be transmitted to further generations.

Session 69

Theatre 3

Genetic determinism of sperm DNA methylation in French Holstein cattle

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Methylation quantitative trait loci (meQTL) are genetic variants that influence DNA methylation. In cattle, little is known about the genetic determinism of DNA methylation, either at the level of individual methylation sites (CpGs) or on a global scale. In this study, we investigated the sperm methylome of 262 Holstein bulls obtained by reduced representation bisulfite sequencing. After filtering out CpGs without individual variability or co-localizing with SNPs, we performed sequence-based GWAS of three groups of methylation phenotypes: (1) principal component scores of the most variable CpG sites, (2) average DNA methylation rate for genomic regions (e.g. whole chromosome, gene, promoter), and (3) DNA methylation rates of individual CpG sites. We then examined the SNPs significantly associated with CpG methylation ($-\log_{10}(P) > 7.3$) and classified them according to their proximity to the CpG, i.e. cis (<1 Mb), cis long-range (>1Mb, on the same chromosome), and trans (on different chromosomes). We found meQTLs for all phenotypic categories, most of which had cis-regulatory effects. In addition, our study identified candidate genes involved in methylation pathways. Taken together, these results demonstrate a genetic determinism of DNA methylation in cattle and thus an interplay between genetic variation and epigenetic regulation. CF is recipient of a CIFRE PhD grant from ANRT and APIS-GENE. This work was part of the POLYPHEME project funded by ANR (ANR-21-CE20-0021) and APIS-GENE.

Genetic evaluations in dairy cattle incorporating an epigenetic source of information

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The extended use of genomic selection over the last decade has doubled the rate of genetic gain in livestock populations, enabling a consistent and sustained enhancement of animal phenotypes associated with economic traits. Nowadays, not only genomic information is available, but intermediary omics features can also be obtained at an affordable price. Here, we simulated the methylation profile of 13,183 genotyped animals using the methylome pattern obtained from 6 dairy cows sequenced with Nanopore Technology. Then, a multiomic model (GOBLUP) was proposed to determine the impact of adding this epigenetic information into genomic evaluations. The heritability recovered by the GOBLUP for the methylation windows agreed to the simulated one for the different scenarios (h^2 of methylation of 0.1, 0.3 and 0.8). Contrarily, the variances for milk yield retrieved by the traditional model, did not capture the effect of the methylation on the phenotype, and thus, the heritability estimate for milk yield captured part of the genetic variance from the methylation genomic windows. The traditional and epigenetic breeding values showed a Pearson correlation of 0.99. Multiomic models may provide a comprehensive decomposition of the heritability estimates by accurately differentiating genetic and epigenetic variance. This enables more informed breeding decisions, optimize selection under specific management or environments that had a differential impact in the animal epigenetics.

Session 69

Theatre 5

Separating genomic imprinting effects and maternal effects in birth weights in Large White pigs

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For a sustainable pork industry, piglets' birth weights play a pivotal role in the survival and homogenous growth of litters. Maternal effects are known to influence this trait. However, the influence of additional epigenetic effects such as genomic imprinting might be of importance. The objective of this study was to disentangle imprinting effects and maternal effects in birth weights of 42,367 piglets with 49,734 individuals in the pedigree. Animal models including sire and dam gametic effects, as well as maternal genetic effects were implemented into Echnidna MMS. Preliminary results revealed a negative genetic correlation between the gametic sire and dam variance of -0.63 and a small positive genetic correlation between gametic dam variance and maternal genetic effects of 0.22. A direct heritability of 0.13 and a maternal heritability of 0.03 was estimated. Additionally, 29.92% of the direct variance could be attributed to genomic imprinting. Although preliminary, this study reveals the occurrence of parent-of-origin effects and separated genomic imprinting effects from maternal effects. This is a first step towards better understanding epigenetic effects that influence the expression of this economically important trait, for future selection decisions.

Exploring the repertoire of regulatory regions on bovine blood cells methylome

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Taking advantage of publicly available CAGE, ATAC- and ChIP-Seq datasets, we aimed at investigating the repertoire of regulatory regions highlighted by bovine blood immune cells methylome. We used reduced representation bisulphite sequencing (RRBS) to profile lymphocytes/CD4, Neutrophils/NT and monocytes methylome. A set of 856,825 common CpGs was retained (48.10% in intron/intergenic regions). ~300k CpGs annotated in intron/intergenic regions colocalizes with CAGE and/or H3K4me3 signal putatively highlighting unannotated transcripts. From ~110k CpGs colocalized with other features than promoter/TSS, a significantly enriched ($p < 10^{-6}$) with enhancer associated states (EAS) was observed. Differentially methylated cytosines (DMCs) were identified using methylKit ($\Delta\text{meth} \geq 50\%$; $\text{padj} < 1\%$) comparing (i) CD4 vs other cell-types and (ii) NT vs other cell-types. ~13k and ~2,9k DMCs were identified from comparisons i and ii, respectively. DMCs were predominantly annotated in promoter regions but also colocalized with EAS. 94.4% and 41.5% of the DMCs colocalized with EAS were indeed hypomethylated (hypo) in NT and CD4 cells, respectively, suggesting active and/or decommissioned enhancers. Interestingly, 12 DMCs hypo in NT colocalizes with regulatory regions of LSP1 gene; expressed in blood immune cells affecting the host response to infection and immune system homeostasis. Our results highlight the importance multi-omic approaches to better understand the functional role of regulatory elements on blood immune cells. Updated results on the functional characterization of cell-subtype specific markers including lymphocytes B, T-CD8, T-CD4 and Natural killers will be shown.

Transmission of sperm DNA methylation patterns to the embryo in cattle: a genome-wide analysis

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After fertilization, DNA methylation patterns in mammals are massively but not totally erased, and the sperm DNA methylation features transmitted to the embryo could be a support for inter-generational epigenetic effects. While bull semen is a commercial product widely used for artificial insemination (AI), little is known about the magnitude of this phenomenon in cattle. We selected four Holstein AI bulls with divergent DNA methylation patterns obtained by reduced representation bisulfite sequencing (RRBS). For each bull, frozen semen was used to generate blastocysts using slaughterhouse ovaries in three independent in vitro fertilization experiments. A total of 14-16 expanded blastocysts collected in the three experiments were then pooled to obtain one biological replicate, and three replicates were obtained per bull. Genomic DNA and RNA were coextracted using the AllPrep Qiagen kit and the DNA eluate was used to generate RRBS libraries. After sequencing and quality control, reads were mapped on the in silico bisulfite converted cattle genome (ARS-UCD1.2 assembly) using Bismark. As expected, all embryo replicates showed genome-wide hypomethylation, but some methylated features in sperm retained a certain degree of DNA methylation in the embryo (CpGs with at least 20% methylation in both sperm and embryo). Using a partial least square discriminant analysis on this subset of CpGs with both sperm and embryo samples (MixOmics R package), we identified 3033 CpGs whose inheritance in the embryo varied according to the bull, suggesting bull interindividual variations in the ability to be reprogrammed or transmit paternal DNA methylation patterns. These CpGs were incorporated to the design of the RUMIGEN EpiChip. ARRS is a recipient of a PhD grant from INRAE PHASE. This project is part of the INRAE DIGIT-BIO program "DINAMIC".

Identifying the rules for defining CpG epigenetic bovine diversity. A study on public RRBS data from sperm isolated from Montbéliarde and Holstein bulls

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Breed epigenetic diversity was recently detected in muscle of pigs and blood of cattle, probably as a result of long-term selection for morphological adaptive and quantitative traits, that persists after embryo epigenetic reprogramming. In our study, the breed epigenetic diversity in the male germline from Holstein (H) and Montbéliarde (M) bulls was investigated using public Reduced Representation Bisulfite Sequencing (RRBS) data, publicly available at the NCBI database. Open source Whole Genome Sequencing data from H and M animals were used to estimate genetic diversity between the two breeds and, thus, correctly assess CpG positions with low frequencies or absence of SNPs. Sperm epigenetic diversity was studied in 321,215 SNP-free CpG positions, for which methylation clearly distinguishes between H and M breeds. A total of 6,099 differentially methylated cytosines (DMCs) were identified. Analyses of pattern of distribution revealed that DMCs: i) were partially associated with genetic variation, ii) were consistent with epigenetic diversity previously observed in bovine blood, iii) presented long-CpG stretches in specific genomic regions, and iv) were enriched in specific repeat elements, such as ERV-LTR transposable elements, ribosomal 5SrRNA, and BTSAT4 Satellites. This study, based on publicly available data from two cattle breeds, contributes to the identification and definition of distinct epigenetic signatures that may have potential implications for mammalian embryo anatomy and development.

Effect of supplementation during the rearing phase on the DNA methylation of *Longissimus thoracis* muscle in F1 Angus-Nellore cattle at weaning

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This study assessed creep-feeding supplementation effects on *Longissimus thoracis* (LT) muscle genome methylation using crossbred cattle. Forty-eight F1 Angus-Nellore non-castrated males were divided into two groups: G1 (n = 24) – no creep-feeding; G2 (n = 24) – creep-feeding. After weaning (210 days), all were feedlot-finished for 180 days under identical conditions. Groups differed significantly (P < 0.05) in weaning weight and LT muscle marbling score (G2 > G1). At weaning, LT muscle biopsies were collected for genomic DNA methylation analysis via reduced representation bisulfite sequencing (RRBS). The G2 showed higher average methylation levels (0.18% more CpG, 0.04% CHG, and 0.03% CHH). 974 regions with differential methylation (DMRs: >25% and q < 0.05) were found, overlapping with 241 differentially methylated genes (DMGs), 108 hypermethylated and 133 hypomethylated in G2. 39 DMGs were previously identified as differentially expressed genes (DEGs: log2 FC [0.5] and FDR 5%). Overrepresentation analysis highlighted epigenetic regulations related to muscle growth, PPAR signaling, adipogenesis, insulin response, and lipid metabolism. These changes improved supplemented animals' performance up to weaning and enhanced meat quality, including greater backfat thickness and intramuscular fat.

Comparison between indicine and taurine cattle DNA methylation patterns across different tissues

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In a previous study, Reduced Representation Bisulphite Sequencing (RRBS) was conducted on blood samples from 5 taurine Angus and 5 indicine Nellore bulls, revealing 25,765 differentially methylated cytosines (DMCs). Most DMCs were concentrated in CpG-rich regions near genes associated with cellular and anatomical morphogenesis not expressed in blood cells, suggesting a tissue-independent initiation of epigenetic marks warranting further investigation. Moreover, sequences surrounding DMCs showed an enrichment of SNPs compared to other CpGs, indicating a potential link between differential methylation and genetic diversity. The EPI-ADAPT project is expanding upon this research, incorporating more animals, tissues, and omics data to explore and compare methylation and gene expression patterns across tissues and subspecies. This endeavor aims to elucidate the epigenetic regulation of genes involved in the differentiation and adaptation of the two subspecies to diverse environments.

Session 70

Theatre 1

Energy Use in the EU Livestock Sector: A Review Recommending Energy Efficiency Measures and Renewable Energy Sources Adoption

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This study reviews a large number of life cycle assessment-based studies covering energy use in major EU livestock systems including dairy, beef, pork, broiler and egg production systems. The reviewed studies indicate a high fossil energy dependency across all livestock systems. Generally, the energy embedded in the production of animal feed emerges as the predominant contributor to energy consumption across most livestock systems, except in beef production systems, accounting for around 50-75% of all energy use. On farm-energy use consumption varies depending on a range of factors but is generally concentrated in animal housing and manure management while depending on the production system electricity-lighting, feeding and milking systems are also shown to be significant energy consumers. This suggests several pathways to reducing fossil dependency within livestock agriculture including through the increased adoption of renewable energy systems, implementation of energy efficiency measures and the adoption of sustainable production practices. As a major limitation, our analysis indicates that considering the size and variation in livestock production systems there is relatively little accurate and detailed data available across the EU and that there is a need for developing a standardized approach to energy measurement in livestock systems as well as the wider agricultural sector which will further support the development of targeted interventions aimed at reducing fossil fuel dependency and mitigating the sector's impact on climate change.

Utilizing energy auditing in intensive livestock farming facilities

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Livestock farming facilities encompass a variety of structures designed to house animals, ensuring their health, welfare and productivity. These facilities can range from traditional barns and pens to more modern intensive housing systems. Key considerations in the design and management of these facilities include ventilation, lighting, temperature control, waste management, as well as access to water and feed. Proper facility design and maintenance are essential for maximizing animal comfort. Livestock farmers often employ a combination of traditional practices and modern technologies to optimize facility performance and ensure the well-being of their animals, while also meeting regulatory requirements and sustainability goals. Energy audits play a crucial role in assessing the energy efficiency and environmental sustainability of these facilities. These audits involve a systematic evaluation of energy consumption patterns, equipment efficiency, building insulation, lighting systems, heating and cooling systems, and renewable energy potential. By identifying areas of inefficiency and opportunities for improvement, energy audits help farmers reduce operating costs, minimize environmental impact and enhance overall farm productivity. Implementing the recommendations from energy audits may involve upgrading equipment, improving insulation, retrofitting lighting systems with energy-efficient LEDs, adopting renewable energy sources such as solar panels and implementing energy management strategies to optimize energy use. Through energy audits and subsequent improvements, livestock farmers can enhance the sustainability and profitability of their farms while also contributing to environmental stewardship efforts.

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Theatre 3

Simulation model for renewable energy systems in livestock barns: three case studies

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The livestock sector contributes to approximately 12% of global greenhouse gas (GHG) emissions. On-farm energy use accounts for 7.6% of total GHG emissions in livestock farming. This is due to heavy reliance on fossil fuels to meet the heat requirements of animals, particularly young ones. Only 4% of this energy is currently sourced from renewable sources. Energy simulation models, both on the supply- and demand side, save resources and time by minimizing trial and error. A model was developed using a quasi-steady state approach. Given electricity and heat loads, the program calculates hourly energy, cash, and emissions flows throughout the entire system. Monthly energy data for 2020 were available for three RES4LIVE pilot farms: pigs (Belgium), laying hens (Greece), dairy (Germany). Their electricity and heating fuel consumption were averaged to hourly values each month. Weather data was sourced from Solcast. Economic and ecological parameters for each country were obtained from Eurostat. Optimal solutions were determined by minimizing life cycle costs (LCC) and fuel GHG emissions over a 20-year period. The pig farm can achieve up to 97% emission reductions, 58% reduction in LCC but with an increased investment cost of 30 times compared to a gas boiler. The dairy barn can achieve 61% emission reduction, accompanied by a 44% reduction in LCC and 38.5 times higher investment cost. In the poultry barn, reducing grid emissions by 89% requires an LCC of 2.08 times the current installation. A trade-off is found by reducing emissions by 45% and LCC by 4%, while increasing the investment cost by four. Variable energy demands and emissions from life cycle analysis have been explored, yielding different results, particularly for electrical batteries.

The AgEnergy platform – a tool to search and assess fossil-energy-free technologies and strategies

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The AgEnergy platform (<https://platform.agrofossilfree.eu/>) is the main result of the H2020 Project AgroFossil-Free that ended in September 2023. Its main objective was to create a framework under which all core stakeholders would cooperate to evaluate and promote the currently available fossil-energy-free technologies and strategies (FEFTS) in EU agriculture to decrease in the short term and eliminate in the long run the use of fossil energy in farming processes, while maintaining yield and quality of the end product offered. Such framework was expected to contribute in bridging the gap between the available novel high-end clean energy solutions and the everyday European agricultural practices by capturing grassroots-level needs and ideas, promoting effective exchange of information and investigating the possible financing opportunities for any de-fossilizing activity in EU farms. Ultimately, it facilitated farmers to find solutions for their specific needs contributing towards fossil-free farming through the platform. AgEnergy Platform is an online and interactive tool for stakeholders to communicate, interact and share knowledge by deploying the collected information and knowledge on FEFTS in the form of easily accessible end-user material. Its design is based on user-friendliness, supporting several levels of filters in order for the end-users to easily identify which FEFTS is required by them. Additionally, it offers the opportunity for assessment of the inventoried FEFTS by the stakeholders, allowing for the crowdsourcing of grassroots-level ideas and needs for research; and integrates an AI Decision Support Tool to propose interventions/FEFTS based on user inputs. The platform in its 2 years of operation has achieved to accumulate more than 450 registered users and over 3000 unique visitors. Currently, the platform contains 1953 FEFTS of all types and it is open to be supplied with more FEFTS by any registered user.

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Theatre 5

Sustainable livestock sector under climate transition

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Current production chain in the agricultural sector directly feeds into climate change, which constitutes a major concern for current livestock systems worldwide. Greenhouse gasses (GHG) and its associated changes in mean climate variables and climate variability affect the animal feed supply chain, the water resources as well as animal health and production. The livestock sector has a central role in food supply and security, as it contributes 15% in the meat, milk and eggs and 31% of global per capita calorie and protein supply, while at the same time the demand is expected to increase, due to population growth. Additionally, the Intergovernmental Panel on Climate Change (IPCC) highlights the substantial contribution of fossil fuel use in the livestock farming, indirect emissions, on farm emissions and post farm emissions and the immediate need for the sector to meet the demands of Paris Agreement. In parallel, there is no one-model size approach to reducing emissions from livestock ecosystem. Each production system, species and location possesses unique characteristics, costs and benefits. The current research highlights the pathway and the peak points of livestock sector towards smart livestock farming for climate mitigation. An overview on the four pillars of sustainable livestock will be presented, corresponding to animal comfort, animal feeding, lowering the overall emissions and adapting to the market needs. Through the recording of this framework, good practices as well as deficiencies in livestock management are identified. Additionally, through a bibliometric analysis, the components of good practices on those four pillars are captured over. Bibliometric analysis imprints is conducted with VOSviewer bibliometric software over web of science database in order to gain a one-stop over-view on current research and to identify both the trends and knowledge gaps on livestock farming adaptation due to climate change. Smart farming technologies corresponding to sensors use and low GHG footprint energy applications are the main trends towards climate adaptation.

An integrated renewable energy system for the de-fossilization of a commercial swine nursery barn

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In swine farming, ensuring optimal environmental conditions within nursery barns is crucial for the health and growth of weaners. Maintaining ideal temperature and humidity levels presents a challenge, especially during both hot and cold seasons. Traditional HVAC systems, while effective, often come with high energy consumption and subsequent greenhouse gas emissions. This study focuses on developing a sustainable heating system, using a pilot case of a nursery barn within a swine farm located in northern Italy, housing 500 sows and 2500 weaners. Our approach involves designing and testing a Renewable Energy Source (RES) system that incorporates bore-hole thermal energy storage (BTES) and photovoltaic thermal (PVT) collectors, integrated with a Dual-Source Heat Pump (DSHP). Additionally, a smart control system has been developed and implemented to monitor RES production, energy usage, and environmental conditions both outdoors and within different sectors of the barn. The collected data were analyzed to determine the optimal settings for the integrated system to maximize energy production and efficiency. The effectiveness of the system in controlling the environmental conditions of the nursery barn was evaluated based on various environmental parameters, with a focus on areas occupied by the animals. The results indicate that a combination of renewable energy sources can be effectively defined and integrated into an automated heating system to meet the specific needs of a swine farm. This demonstrates the viability of a project tailored to harnessing renewable resources commonly found in agricultural settings.

Heat Pump HVAC System as Part of an Integrated RES System for the Indoor Climate Control of a Laying Hen House

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Lately, much of the academic community's research efforts have been directed towards the transition from fossil fuel dependence in European husbandry facilities to fostering a sustainable livestock sector. To this end and in the framework of the H2020 RES4LIVE project (GA No. 101000785), a study investigation is conducted in an experimental laying hens facility at the Agricultural University of Athens (AUA), to evaluate the use of an integrated Renewable Energy Source (RES) instead of fossil fuel, while enhancing animal welfare and productivity. The core of the system consists of a heat pump (HP) designed primarily to condition the indoor air by heating, cooling, and dehumidifying it, combined with a solar photovoltaic (PV) system to generate electricity, covering part of the heat pump's consumption. The present study focuses on the results obtained during the testing and monitoring of the innovative HP using a sensor-based monitoring system that collects data on system performance and indoor conditions. Focusing on two particular periods, summer and winter, the initial findings suggest that the HP technology seems to present the potential to enhance animal welfare by maintaining suitable indoor air temperatures and relative humidity into the desired range, especially during heatwaves where the previous conventional heating and ventilation systems were considerably inadequate. The HP achieves a Coefficient of Performance (COP) in the range of 2.5 to 3.5 during summer (cooling), and 3.0 to 4.7 during winter (heating), respectively. The results contribute to expanding our understanding of incorporating RES into livestock systems.

Geothermal energy concepts for livestock applications, analysis of energy savings, life cycle cost and animal welfare

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Geothermal energy provides sustainable thermal energy, it offers many applications in the building sector resulting in optimal comfort with minimal energy cost. The potential for this technology is strongly related to applications where there is both heat and cold demand. This does not seem immediately applicable to livestock applications, with at first sight only heat demand. Yet the addition of cooling offers additional benefits, especially in terms of optimal comfort for the animals and product quality. In addition, geothermal cooling can be offered at a fraction of the energy cost compared to active compression cooling. This paper gives an overview of the possibilities and limitations of using geothermal energy (both open and closed systems, type ATES or BTES) for livestock applications. An ATES application uses an underground aquifer, in which groundwater is extracted and injected after changing heat with the stables. This creates an underground hot and cold bubble of groundwater that is built up and down through the seasons. In a BTES application, vertical probes are inserted into the soil, in which a fluid circulates that transfers the heat/cold to the soil. The energy concepts are based on the use of ground-coupled heat pumps combined with passive cooling. The analysis is based on some concrete cases in the EU project RES4LIVE. An overview is given of the energy concepts, energy consumption, environmental impact and comfort benefits. In addition, the total cost of ownership is examined with a life cycle cost analysis and dynamic payback calculation. The application of geothermal energy requires special attention to the different climatic conditions for different European countries. This will create variable operating conditions in North, Central and South Europe which will result in a different economic and ecological impact.

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Theatre 9

Life Cycle Impact Assessment of an Integrated PVT-BTES-Heat Pump System for a Commercial Swine Farm in Italy

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This study performed a Life Cycle Impact Assessment (LCIA) of an innovative energy system deployed at a commercial swine farm in Italy. The integrated Renewable Energy Sources (RES) system combines Photovoltaic-Thermal (PVT) panels, Borehole Thermal Energy Storage (BTES), and a Heat Pump (HP) to provide heating to the farm's nursery barn, where 13,500 weaners are produced annually. The system is designed to harness solar energy through PVT panels, simultaneously generating electricity and heat. Excess thermal energy during summer is stored in the BTES for use during the winter period when the HP efficiently exploits the thermal energy of the outdoor air and/or the BTES. The LCIA considered the raw material extraction, manufacturing, transportation, and installation phases for each system's components separately. The environmental load for all PVT, HP and BTES components was expressed per 1 kg of weaned piglets at the gate of the nursery barn. Primary data, such as bills of materials, were collected from the farm and technology providers. International secondary datasets and literature were used to model the upstream processes. The Environmental Footprint (v3.1) methodology as available in the SimaPro 9.5 software was implemented for estimating the environmental impact category indicators. The use of fossil fuels (RUF) and climate change (CC) were identified within the impact categories connected with higher indicator estimates per kg of weaned piglets at the farm gate for all PVT (RUF: 0.03MJ; CC: 2.14g CO₂eq), BTES (RUF: 0.004MJ; CC: 0.26g CO₂eq) and HP (RUF: 0.004MJ; CC: 0.28g CO₂eq) technologies. These results will be used to investigate the potential environmental performance improvement of weaned piglets production which is associated with the installation of this integrated RES system.

Modeling of thermal status at animal level – Swine and Cattle: A Review

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Providing proper thermal environment for swine and cattle is a challenge not only in hot climates, but also in temperate climate due to more frequent occurrences of heat waves occurred in recent years. It is thus important to model the thermal status at animal level so the precautions taken to cool animals down can be implemented on time. The aim of this study is to systematically review the modelling approaches of thermal status at animal level for swine and cattle, supported by RES4LIVE project. The database ‘web of science’ and ‘scopus’ were used for literature search. The literature search was conducted by combining keywords (‘swine’ OR ‘pig’ OR ‘sow’ OR ‘cattle’ OR ‘cow’) AND keywords ‘modeling’ AND abstract (‘thermal’ OR ‘heat’). Language was limited to English. There were 186 papers found in ‘web of science’ and 284 found in ‘scopus’, within which 28 duplicated articles were removed. The review was conducted by following a review protocol of ‘ROSES’ and was screened following the order of title, abstract and full paper. The review indicated that three modelling methods are found, including semi-experimental model, mechanistic models, and machine learning. The semi-experimental model was relatively easy to be used but formulated per given livestock production buildings and is thus difficult to be widely applied. Mechanistic models can be generalized but require knowledge of heat transfer and input of some parameters obtained from limited data can bring uncertainties. Machine learning is becoming popular, but physics constrains are needed for reasonable predictions. It is critical to consider physiological regulations in modelling and individual differences in adapting to given thermal conditions as well.

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Theatre 11

The effect of renewable energy transition on the thermal comfort in a pig farm – animal compartment level experimental investigation

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Renewable energy integration in livestock farming is crucial for curbing CO₂ emissions, with on-farm energy consumption being a significant contributor. The RES4LIVE project, funded by the EU Horizon 2020 program, aims to diminish fossil fuel dependency in livestock farms. As part of this project, a modular heat pump was installed at a farrow-to-finish pig farm in Melle, Belgium. The system consists of a low and a high temperature heat pump with an overall heating capacity of 60 kW. The primary heat source for this system is thermal energy from photovoltaic thermal collectors (PVT). In total, there are 24 flat plate PVT panels installed at the farm, offering an electrical capacity of 8.40 kW and a thermal capacity of 32.8 kW. The integrated installation of the modular heat pump and the PVT panels is planned to deliver the whole heating demand of the farm (220 MWh per year), and replaces an existing gas boiler. Thermal energy from the heat pump is conveyed to the farm using water as a heat transfer medium. This water circulates through a network of pipes, distributing heat to various systems such as sanitary water lines, air heaters, and underfloor heating. Notably, the air heater lines and underfloor heating play a crucial role in heating the animal compartments. In this study, an examination is conducted on the temperatures within the animal compartments when the heat pump serves as the primary heating source, compared to the gas boiler. The data encompasses 16 fattening, 2 farrowing, and 3 weaned pig compartments, with their respective temperatures measured by Pt1000 sensors and logged every 10 minutes. The operational period of the heat pump spans from January 25th to February 5th, 2024, forming the basis of this experimental investigation. The findings underscore the effectiveness of the integrated heat pump and PVT system in reducing carbon emissions and offering a viable alternative to fossil fuel-based gas boilers on the farm.

Compact Bio-CNG farm filling fuel production from anaerobic digestion: Hollow fibre permeation and Hybrid compression for technically and economically feasible small scale biofuel production

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As several studies have shown, agricultural residues can produce the total fuel requirement for the agricultural sector as non-fossil fuels. Challenges that could be solved largely with the realized pilot plant within this study were the economic feasibility of very small Bio-CNG production plants from 10m³ raw biogas/h and the achievement of fuel quality >97% CH₄. The study shows the results from the operation of the two pilot plants (i) the one with 10 m³/h of raw biogas at the Leibniz Innovation Farm near Potsdam, Germany, and (ii) the other with 35 m³/h of raw biogas in Utzenaich, Austria. The pilot plant in Potsdam has operated since January 2024 and supplies the farm's fuel for CNG-powered tractors. The study describes the measured key performance indicators of the two pilot plants, such as bio-CNG quality, specific energy consumption, and manufacturing costs of market-ready plants. Taking into account current market prices for raw biogas and electricity as well as investment costs for the Bio-CNG plant, total costs for Bio-CNG production (@>200bar) of 1.51€/kg Bio-CNG for the 35 m³/h plant, and 2.25€/kg Bio-CNG for the 10 m³/h plant could be determined. The energy consumption was determined at ~1 kWh per Nm³ of biomethane (purified and highly compressed) in pilot operation. With the results from the monitoring of the two realized pilot plants, the study also determines the expected unit costs per plant as well as specific costs of €/kg Bio-CNG fuel for further plant sizes of 65 m³/h and 100 m³/h raw biogas. The expected cost depression is seen between the system sizes. The development of the small-scale Bio-CNG farm filling station is proving to be technically and economically feasible based on pilot plant data.

Session 70

Poster 13

Air temperature measured by embedded sensors adopted by pig transporting vehicles in a scaled compartment model

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To regulate fans and shutter openings of pig transport vehicles highly depends on the monitoring of indoor air temperature by embedded sensors to ascertain the proper indoor air temperature. It was observed in previous measurements conducted in a weaner transport vehicle that the temperature difference of air (ΔT_{air}) measured by embedded sensors and thermocouples installed by researchers could be up to 20 °C. The objectives of this study are (1) to study if the current installation manner of embedded sensors in lorry and trailer can reflect the air temperature inside compartments, and (2) to propose feasible approaches to install embedded sensors. A scaled compartment model with dimensions of 122 cm (L) x 122 cm (W) x 70 cm (H) was constructed. The length and width were half of the dimensions of a full compartment and the height was maintained the same as the one of a full compartment. Experimental scenarios mimicking winter and summer conditions were tested. The hypothesis was that ΔT_{air} could be reduced by insulating the walls near embedded sensors or by forcing the indoor air to pass over the embedded sensor as much as possible. The results showed that insulation had limited influence on ΔT_{air} for both winter and summer scenarios although insulations could increase the surface temperature of surrounding walls by about 6.0 °C. Forcing the indoor air of the scaled compartment to pass over the embedded sensors could reduce ΔT_{air} to be less than 0.5 ~ 1.0 °C.

Weighted SNP enhances the genomic prediction accuracy for milk citrate predicted by milk mid-infrared spectra of Holstein cows in early lactation

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Previous studies have shown that milk citrate predicted by milk mid-infrared spectra is strongly affected by a few genomic regions. The purpose of this study is to explore whether giving weight to SNP increases the genome prediction (GP) accuracy for predicted milk citrate of Holstein cows in early lactation. A total of 134,517 test-day predicted milk citrate collected within the first 50 days in milk on 52,198 Holstein cows from the first 5 parities were used. There were 122,218 animals in the pedigree, and 4,479 of them had 566,170 SNPs. 181 youngest cows with genomic and phenotypic information were selected as a validation population. We created partial (181 cows without phenotypic) and whole datasets. (genomic) estimated breeding values (EBV or GEBV) in the partial and whole datasets estimated by best linear unbiased prediction (BLUP) with pedigree (ABLUP), single-step genomic BLUP (ssGBLUP, pedigree combine genomic), weighted ssGBLUP (WssGBLUP, pedigree combine genomic using SNP-weighted), respectively. Linear regression method was used to compare EBV (or GEBV) from partial and whole datasets. The prediction accuracies for ABLUP and ssGBLUP were 0.42 and 0.70. The prediction accuracies of WssGBLUP in the 5 iterations with different CT values (determines departure from normality for SNP effects) ranged from 0.70 to 0.86. Our results showed that WssGBLUP is beneficial in improving the prediction accuracy of GP for predicted milk citrate. In addition, our results provide evidence that employing WssGBLUP for GP accuracy is beneficial for traits that are highly affected by a few genomic regions.

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Theatre 2

Estimation of heritabilities and genetic correlations in very large datasets using predictivities within and across traits

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Under genomic selection, heritabilities for strongly selected traits are reduced, and antagonism between production and fitness traits can intensify. However, estimating genetic parameters by established methods such as REML or Bayesian with large genomic data is computationally expensive or unfeasible. On top of it, these methods estimate parameters for the base generation, but those from the current generation are more suitable for use in the selection index. In this study, we investigated using theoretical and realized accuracies to estimate heritabilities and predictivity across traits to estimate genetic correlations. The new methods require GEBV derived from all but the latest generation and adjusted phenotypes for genotyped animals in the last generation. Testing included dairy, beef, and pig datasets. The largest dataset included 18-type traits of 5M Holsteins with about 700k genotyped animals. For comparisons, estimates were obtained from time slices by REML using genotypes when computationally feasible. Preliminary results indicate that the estimates by predictivities are realistic with large data sets when genotyping becomes widespread rather than selective. Heritabilities estimated by predictivities were generally lower than those estimated by REML. However, estimates by REML using samples of data slices with incomplete or missing genotypes due to high computing costs are likely biased. Parameter estimation by predictivities is technically applicable for large datasets; however, more research is needed to evaluate their properties.

Multi-breed multi-trait single-step genomic predictions for Holstein and Jersey including crossbred animals
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Genomic selection for crossbred animals is challenging due to difficulties in establishing suitable multi-breed reference populations and modelling missing pedigree information. We investigated the benefits of multi-breed multi-trait single-step genomic evaluations that jointly analyse New Zealand data from two purebred populations (PBD, Holstein and Jersey) and a derived crossbred population (XBD). We also investigated the impact of modelling missing pedigree information using genetic groups (GG) or metafounders (MF). Pedigree, genotypes (127K), and phenotypes for 305-days milk yield (MY, 538K) and calving season days (deviation between planned and actual calving date, CSD, 370K) were available for PBD and XBD cows. Six scenarios were implemented with MY and CSD jointly analysed in a multi-trait model: A) a single-step evaluation per breed, each using phenotypes of all breeds treated as a single trait, but only genotypes of the respective breed, and 255 GG; B) a joint evaluation using the genotypes of all breeds, with phenotypes and GG as in A; C) as B but replacing all GG by MF; D) as B but replacing all GG by 4 MF; E) as B but grouping all GG into 4 GG; F) as B but with phenotypes from different breeds treated as separate correlated traits. Using all PBD and XBD genotypes gave higher accuracies than analysing each breed separately. Scenario F showed the highest accuracy among all scenarios for MY but the lowest for CSD. Reducing the number of GG gave similar results to using all GG, while using MF had limited benefits. Combining Holstein, Jersey, and derived XBD data into multi-breed single-step evaluations can enhance the accuracy of genomic predictions for PBD and XBD animals.

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Theatre 4

Over-time genetic correlations based on accuracy in GEBV for 18 linear type traits in US Holsteins

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Genetic parameters change over time in populations under genomic selection. Unbiased parameters can be estimated if all phenotypes, pedigree, and genotypes are available. However, the estimation process is challenging or even impossible with many genotypes. This study aimed to apply formulas based on accuracy (acc) of GEBV to compute genetic correlation and heritability. Heritability (h^2) was computed as $(c^2 + \sqrt{(c^4 + 4c^2Me/N)})/2$, where c is predictivity = $\text{Corr}(u+e, \hat{u})$; \hat{u} = vector of predicted breeding values, Me = number of independent chromosome segments (15,000), and N = number of genotyped animals with phenotypes. Genetic correlations (rg_{ij}) between traits i and j were computed as $\text{Corr}(u_i + e_i, u_j)/(h_i \text{acc}_j)$, where $\text{acc} = c/h$. With 4.9M phenotypes from 2001 to 2023 from the national evaluation data from Holstein Association USA, a single-trait model was used to obtain GEBV for 18 traits with 6 validation datasets in every two years from 2009 to 2020. Training datasets ended before the first validation year. Genotypes were available for 366K bulls and 326K phenotyped cows. Correlations (rg) were compared with those from variance component estimation (VCE) via Gibbs sampler and Pearson correlations (rp) among GEBV. The rg were like from rp but lower than those with VCE. Some differences could be due to the base population in VCE and the last generation in rg and rp depending on given h^2 . The rg could be close to true parameters after adjustment by acc. Rank correlations of GEBV with rp and rg were high, 0.99, over time, indicating good concordance. With the proposed formulas, computing time for genetic parameters in large populations can be drastically reduced; however, rescaling the estimates may be required.

Genome-wide association study for stature for Holstein cattle based on whole genome sequence, using a mixed linear model

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Genome-wide association studies are one of the most popular tools for detecting genetic variants associated with variation of quantitative traits. The goal of our study was to identify significant SNPs and genes associated with stature based on the whole genome sequence information of 532 Holstein bulls from the 1000 Bulls Genome Project, as well as the estimated breeding values and pedigree data available from the Multiple Across Country Interbull Evaluation. Due to the fact that genomic data is large, the number of SNPs was reduced by linkage disequilibrium (LD). A mixed linear SNP-BLUP model was applied to estimate the additive effects of all SNPs, representing the genome-wide set of polymorphisms, using the MiXB LUP 3.0 software. Significant SNPs were selected on the basis of the Wald test. In further steps, significant SNPs were genomically assigned to genes, and genes were functionally assigned to gene ontology terms and KEGG pathways. The last step of the analysis was the enrichment analysis of gene ontologies and pathways represented by significant SNPs.

Exploring dynamics of breeding values under non-random mating and imprinting: A real example with Rubia Gallega breeding program

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Breeding value, originally framed within the Hardy-Weinberg equilibrium, faces challenges in livestock populations characterized by selective selection pressures and non-random mating. Consequently, understanding the dynamics of breeding value under these settings is important. For example, imprinting and different allele frequencies between sexes introduce a different variance contribution of each sex, along with the emergence of new variance terms. This study aims to establish an understanding of breeding values within the context of sex-specific mating as compared to random mating. Specifically, under sex-specific mating, breeding values for males and females depend on the allele frequencies of each sex and on the presence of imprinting. These concepts were studied in a dataset from a population of Rubia Gallega beef cattle. The study identified multiple genomic regions exhibiting differential allele frequencies between reproductive males and females. Furthermore, the results showed discrepancies between breeding values calculated under random mating on the one hand, and sex-specific mating on the other, potentially leading to shifts in ranking both within and between sexes.

Genomic evaluation of litter size variability in Large White pigs

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The objective of this study was to analyse the genomic background of litter size variability. We used 199,520 observations on total number born (TNB) from 46,901 Large White sows. Firstly, the variance components and heritability were estimated for TNB using ASReml 4.2. Then the log-transformed variance of residuals of TNB (LnVar) was estimated to obtain the phenotypic variability of litter size and its variance components. Finally, the LnVar was used in a genome-wide association study (GWAS) with a whole-genome sequence data of 1,570,396 SNPs genotyped on chromosome 7. The GWAS was run in PLINK 1.9 with the mixed model. The results of post-GWAS on chromosome 7 indicated that out of 30 most significant SNPs, four were in a close proximity to potential candidate genes: DAAM2, FARS2, F13A1, AKAP9. Those genes were found to be associated with reproduction in different species. Further analysis of the remaining chromosomes is needed to verify additional potential candidate genes associated with litter size variability.

Effect of the number of sibs measured in commercial conditions on the response to selection for body weight in turbot

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The turbot (*Scophthalmus maximus*) stands out among aquaculture species due to its high commercial value. Presently, the main breeding goal in selective programs for this species is body weight. However, selecting directly on weight recorded solely in the nucleus (NW) is inefficient to improve commercial weight (CW) given the distinct husbandry conditions between environments and the existence of genotype-by-environment interaction. Therefore, to effectively improve CW, it is necessary to base selection decisions on records of relatives (e.g., sibs) of the nucleus candidates reared in the commercial environment. For traits recorded on relatives, genomic evaluation has shown higher accuracy and genetic gain compared to pedigree-based BLUP but the number of sibs required to be recorded and genotyped remains uncertain. This simulation study aims to determine the optimum number of weighed and genotyped sibs under commercial conditions to ensure accurate estimates of the breeding values for CW in the nucleus candidates. A simulated base population of 22,000 individuals was generated using empirical genotypes of 5,000 individuals for a low-density 5K-SNP chip. Two traits, NW and CW, were simulated with heritabilities of 0.30 and a genetic correlation of 0.5 between them. The nucleus comprised 100 full-sib families with 20 candidates per family. Five different scenarios varying in the number of sibs recorded and genotyped in commercial conditions were compared. After 3 generations of selection, increasing the number of sibs from 10 to 20, 30, 40 and 50 increases accuracy and response to selection by approximately 6%, 9%, 11% and 11%, respectively. Consequently, it is anticipated that there are no advantages to further increasing the number of sibs beyond 40.

Single-step genomic BLUP breeding value estimation in multi population Atlantic Salmon data

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The study aimed to investigate the impact of corrections for the base-population differences between the A and G relationship matrices with and without group contributions in single-step genomic BLUP (SSGBLUP) models for a multi-population and multi-generation harvest weight data set on an Atlantic salmon breeding population. We also assessed how incorporating J values and Q group contributions into the SSGBLUP models influences the genetic trend estimates over multiple generations. The dataset comprised four populations classified by year classes, each extending over six generations. It encompassed 47,596 harvest weight records and pedigree data for 61,927 fish across seven generations, along with genotype information for 22,899 fish. Various SSGBLUP models and pedigree-based BLUP predictions, with or without genetic groups or J factor, were investigated using a single-trait model. The comparison of the different SSGBLUP models were carried out by estimating level biases, inflations, and ratios of accuracies of GEBV using the Legarra-Reverter method. Results showed that incorporating J factor and groups into SSGBLUP models, whether as random or fixed covariates, enables the estimation of genetic trends across generations with marginal improvement in ratio of accuracies and maintains low inflation biases, although with a slight increase in level bias. Implicit incorporation of the J factor as a random covariate resulted in reduced level biases when compared to fitting J factor as fixed covariate, suggesting the preference for SSGBLUP models with J factor as random covariate alongside genetic group effects.

Session 71

Theatre 10

Supporting the implementation of genomic selection in a guide dogs' population using simulation

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For service dog populations, applying genomic selection would enable more efficient breeding for complex traits such as health, welfare, and trainability. However, the transition from phenotypic to genomic selection requires genomic information. Different data collection scenarios can be envisioned based on the number of individuals, the number of markers, and the genotyping technology. The aim of this study was to identify the optimal scenario for data collection to implement genomic selection and investigate complex trait architecture with whole genome sequence (WGS) information. To do so, we simulated the UK Guide Dogs' population based on their pedigree and existing high-coverage WGS data with AlphaSimR, and then phased and imputed with AlphaPeel for various scenarios. The existing pedigree was extended with additional generations to evaluate scenarios' outcomes in the future. The scenarios considered were composed of diverse genotyping densities and sequencing coverages for the puppies. All scenarios were compared using individual imputation accuracy against the true simulated WGS. Low-pass sequencing scenarios (0.5 to 2X depth) achieved accuracy of 0.986 to 0.998. SNP array genotyping (25K to 710K markers) was inferior, with an accuracy of 0.560 to 0.732. For the UK Guide Dogs, the simulation revealed low-pass sequencing as the best strategy for obtaining WGS information for downstream use in genomic selection and analysis of complex traits.

Integrating foreign estimated breeding values into the national single-step genomic evaluation of German beef cattle breeds

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The Interbeef evaluation estimates breeding values across 16 countries. We define and validate an approach for integrating international pedigree-based EBVs into single-step national evaluations of four German beef cattle populations (Angus (DA), Charolais (CHA), Limousin (LIM) and Simmental (SIM)). The national data were obtained from the beef cattle evaluation of December 2022. Direct and maternal genetic effects for Adjusted Weaning Weights (EBVINT) and associated reliabilities were obtained from the Interbeef evaluation of January 2023. 587,889 Interbeef breeding values were available across the four breeds. To evaluate the impact of integrating foreign data on the national evaluation, we compared the genomic enhanced breeding values (GEBV) obtained from the national evaluation blended with foreign data () with those obtained from a national evaluation without foreign data (). The degression of EBV reverses the regression process to obtain the pseudo-phenotype data for the animals with their own phenotype records. For an unbiased deregression, the expected Pearson correlation between the EBV obtained before and after the deregression is 1. Compared to the scalar deregression approaches, the matrix deregression approach showed a higher correlation to the initial EBV. The results from our study show an approach to include foreign information in the national single-step evaluation for 200-DW and Mat-effect, which reduces bias in national beef cattle evaluations.

Session 71

Poster 12

The effect of correlated herd effects on the variance component estimate

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Herd effects are an important element to consider in livestock genetic evaluations, as differences in climate and management practices influence production outcomes. Often, herd effects are incorporated into the effect of the contemporary group, typically a herd-year-season combination. When modelled as random, these herd effects, which reflect part of the environmental factors affecting a phenotype, are usually assumed as independent. It is, however, reasonable to assume that herds in geographical proximity have some level of correlation in their effects, as they share similar climates and may adopt similar management practices due to social customs. Previous studies have shown that accounting for spatial relationships between herds in smallholder breeding programs can successfully model correlated herd effects. These studies used data spanning a short time period, and when attempting to extend the use of spatial correlations between herds to data sets comprising a large time span, we observed an explosion in the variance component estimates of herd effects. The most likely reason for such an explosion in variance is that, when herds present many years of data, an intra-herd correlation is built-up, and different herds may have different intra-herd correlation levels. Spatial correlations ignore this longitudinal component, and we observed that the consequence of assuming inter-herd correlations without distinguishing intra-herd correlations is a greatly over-estimated variance component for the correlated herd effects. Thus, the longitudinal component must also feature in the spatial correlation matrix to adequately model herd effects.

Exploring Genetic Trends in Elite Canadian Rams

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Genetic gain in the Canadian sheep industry has increased profitability over the past 10 years. Among the traits currently evaluated in the national breeding program are lamb survival (SURV), birth weight (BWT), adjusted 50 days weight (ADJW50), gain 50-100 days (GAIN), loin thickness (LOIN), and fat thickness (FAT). The main goal of this study was to explore the genetic trends in the population based on all animals with records and compare these trends to the ones observed for the elite rams. In addition, to verify the coincidence of ranking of the elite rams for the traits, the rank correlation was estimated across traits. Canada's national sheep evaluation combines all pure and composites in a model using a genetic group strategy to address the effects of individual breeds or breed groups. Data included traits recorded on 706,074 animals (2012-2022), rams with at least 10 progenies by 4,223 rams. Breeding values were estimated by Best Linear Unbiased Prediction (BLUP) using an animal model in the MiXBLUP software. Rams were ranked for each trait, and the top 1% and 10% rams were used to estimate the rank correlation across traits and the genetic trends for the elite rams. Genetic gains were estimated by regressing estimated breeding values of traits on year of birth of the animals and then the gains were expressed as units of additive standard deviations of each trait. Rank correlations ranged from -0.03 to 0.04 for the top 1%; and from -0.23 to 0.13 for the top 10% rams. The standardized genetic gains estimated for the population and top 1% rams were 0.04 and 0.03 (SURV); 0.05 and -0.02 (BWT); 0.05 and 0.04 (ADJW50); 0.09 and 0.01 (GAIN); 0.02 and 0.03 (LOIN); and 0.02 and -0.02 (FAT); respectively. Moreover, the breeding values for the top 1% sires were, on average, two to three folds higher than the average population. Our results suggest that most genetic trends were in a favorable direction, indicating genetic progress for all the traits over the years. The low rank correlation observed for the top 1% and 10% sires highlights the importance of using a selection index to make optimal selection decisions.

Session 71

Poster 14

Impact of season and genetic line on honeybee gut microbial composition

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In this study, we aim at deepening the knowledge on how host genetics and environment affect the gut microbial composition of honeybees. Seventy-seven colonies from eight maternal genetic lines entered this study, these were from a breeding population located in Lombardy, Italy. Worker bees were sampled in three different times of the year 2021, i.e. June, July and October. From each beehive, gut from ten worker bees were collected. The V3-V4 region of the 16S rRNA gene was amplified and sequenced. Colonies were phenotyped for honey yield and docility. The statistical analyses included testing the impact of genetic lines, month of sampling and their interaction. All the bioinformatic and statistical analyses were performed in the R environment. Results showed differences between in honey yield and docility between the some of the genetic lines. For the α -diversity, the ANOVA performed on different indices revealed a significant and consistent impact of month of sampling, while the effect of the genetic line was significant on the Shannon and Simpson indices only. Whilst not significantly different, two colonies shows a tendency to have lower values of diversity. For the β -diversity, PERMANOVA revealed the months of sampling as the strongest effect, the genetic line effect was barely significant, and their interaction was not. The season of sampling, as an indicator of bees' diet, is confirmed to be the major factor. Host genetic line show a weak effect, but the use of queen's genomic information might help in estimating this effect.

Utilisation of dense longitudinal records from automatic milking to assess heat stress effects in dairy cows
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Automatic milking systems (AMS) generate a large number of traits and parameters in dense intervals, which can be used to accurately assess cow behaviour and responses to environmental stressors (e.g., heat stress). In this regard, 188,383 records from cow visits in AMS were merged with temperature-humidity-indices (THI). For the assessment of time-lagged and prompt THI effects on longitudinal AMS traits, the average daily THI was calculated for different periods reflecting the distance (in days) previous to the respective recording date in AMS: THI_0d, THI_1-3d, THI_1-7d, THI_8-14d, THI_15-21d and THI_22-28d. AMS traits included milk yield per (DMY) and per visit (MY), fat and protein content per visit (F%, P%), milking time (MTIME), milking interval (MINT), milk yield per minute (MYPM), scan time (STIME) and connecting time of the milking device (CTIME), number of daily AMS visits (NVISIT) and somatic cell score (SCS). Linear mixed models simultaneously considered the effects of days in milk (time dependent covariate) and of THI via regressions on Legendre polynomials of order 4, in consecutive runs for the different THI recording periods. Environmental sensitivity in terms of significantly declining NVISIT and DMY was observed at THI 70. For all THI periods, a decline in F%, P% and increasing SCS was already detected at THI 40. Hence, trait responses from specific AMS visits combined with dense on-farm meteorological measurements can be used as an early-warning system for heat stress, and as functions for heat tolerance genetic evaluations.

Genotype by environment interactions for carcass traits in Icelandic sheep

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The robustness of animal performance across production systems and environments is important for planning a joint breeding program across different environments. Average carcass weight (CW) of Icelandic lambs varies considerably between flocks because of different farming practices and different quality of pasture. The aim of this study was to find if there is genotype by environment interaction for carcass quality in Icelandic lambs. We used slaughter records including CW, EUROP carcass conformation (CC), and carcass fat (CF) of 405,839 lambs from 60 commercial flocks. We tested three models for genetic evaluation of CC and CF, a reaction norm model (RNM) with solutions for contemporary group effects on CW as environmental covariable, a random regression model (RRM) with random regression on individual CW, and a base model assuming no genotype by environment interactions. For both traits, we identified additive genetic variation related to the random regression and reaction norm slope. This represents additive genetic variation for robustness. For validating the predicted breeding values (PBV) for robustness, we compared PBV for 147 rams used in artificial inseminations, calculated with data from 30 flocks, to the realized slope within half-sib groups calculated in the remaining flocks, and vice versa. The correlation was 0.11 and 0.18 for CC, and 0.14 and 0.37 for CF. Using RRM, the correlation was 0.05 and 0.25 for CC, and 0.33 and 0.45 for CF. The accuracy of lamb PBV for CC and CF adjusted for CW was similar or lower using RNM or RRM, compared to the base model. The results confirmed genotype by environment interactions for carcass traits in Icelandic lambs, represented by genetic variation for robustness. However, accounting for genotype by environment interactions did not improve PBV accuracy.

Identifying candidate variants for cumulative egg number by analyzing 1005 fully sequenced layers
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Identifying genomic variants is important to understand the biological background of egg production in layer breeding. We performed a genome-wide association analysis (GWAS) and a transcriptome-wide association analysis (TWAS) to detect candidate genes for cumulative egg number. Beijing-You chickens and White Leghorns were used to generate purebreds (YY, WW) and crossbreds (WY, YW). Cumulative egg number and egg number at 300, 400, 500, 600, and 700 days of age were computed using individual egg-laying recordings. The genomes of 211 WW, 240 WY, 268 YY, and 286 YW were sequenced at ~15× coverage. After quality control 934 animals, and 12,495,895 SNPs were available for subsequent analyses. A single SNP GWAS was performed using WOM-BAT and additive and additive-dominance models. At 22, 35, 46, 72, and 100 wk, the ovary from six chickens of each genetic group was collected for RNA sequencing. MatrixeQTL and S-PrediXcan were used to perform eQTL mapping and TWAS analysis, respectively. Results showed no significant variants using the additive model. By contrast, the additive-dominance model identified 2,603 additive SNPs and 2,969 dominance SNPs, which were located on chr 1, 2, 3, 6 and 13 and especially affected egg production during the late laying stage. Using GWAS summary data from the additive-dominance model and cis-regulated eQTL, we identified 359 candidate genes for egg production. These results suggest that the additive-dominance model is more powerful to detect the variants for egg production traits of layers in a mixed population with purebred and crossbred animals. Moreover, multi-omics data allows deciphering genetic mechanisms underlying complex traits.

Session 71

Poster 18

Using CADD scores as prior information to improve genomic prediction accuracy in pigs and mice

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In genomic prediction, SNPs contribute differently to estimated breeding values. For instance, some SNPs may be very informative as they are (linked to) causal variants, while others provide no information. Combined Annotation Dependent Depletion (CADD) scores integrate a variety of genomic features to evaluate the likely impact of genomic variants on any phenotype, suggesting that CADD scores can potentially be used to detect important SNPs linked to causal variants. Although previous studies did not show a consistent improvement of accuracy, many different ways of using CADD scores have not been investigated yet. Our objective is to explore different applications of CADD scores to process SNPs, including pre-selecting, classifying, and weighing. Here we will use CADD scores to prioritize SNPs in pigs and mice. Populations will be analyzed separately following three steps. First, SNPs will be selected based on varying thresholds of CADD scores, with threshold sets as >0, 1, 10, 20, or 30. Second, two scenarios will be considered: (1) only using SNPs selected based on these thresholds, or (2) using both selected SNPs and unselected SNPs, separated into two categories. For scenario (1), we will use regular GBLUP. For scenario (2), we will use two modelling approaches: a. BayesRC which uses two groups of SNPs (selected and unselected), or b. GBLUP with two GRMs constructed using two groups of SNPs (2-GBLUP). For the GBLUP models, we will include variations of the model where scaled CADD scores will be used as weight factors to construct the GRM. We will compare the accuracy of all methods with regular (unweighted) GBLUP and 2-GBLUP. Our study will enhance understanding of using CADD scores to improve genomic predictions.

Survival Analysis of Iranian Holstein Dairy Cattle with BLUP and ssGBLUP Using Random Regression Model
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The objective of this study was to estimate the genetic parameters of longevity up to 100 months after first calving with best linear unbiased prediction (BLUP) and single-step genomic BLUP (ssGBLUP) using random regression (RR) model. Data were supplied by the Iranian National Animal Breeding Center which were 30,947,911 test day records from 1,363,534 animals in 4,370 herds and 2419 genotyped bulls. Different scenarios evaluated to determine the best combinations of scaling factors (τ for inverse of the genomic matrix and ω for inverse of the pedigree matrix of genotyped animals). The best method of genetic evaluation of survival determined according to the validation metrics, accuracy and dispersion of predictions. The average of survival from first calving to the last test date was 37.5 month. The heritability of longevity up to 100 months after first calving was estimated to be 4.9%. The accuracy rate (and the dispersion of predictions) in RR-BLUP and RR-ssGBLUP models with default values of $\tau=\omega=1$ were 0.47 (1.63) and 0.53 (0.93), respectively. In the RR-ssGBLUP model, with values of $\tau=1$ and $\omega=1.3$, the accuracy rate (and dispersion of predictions) was 0.54 (0.99), respectively. The results indicated that RR-ssGBLUP model, using values of $\tau=1$ and $\omega=1.3$ can be recommended for the genetic evaluation of longevity.

Implementation of longevity genetic index in Italian Jersey

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The Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana (ANAFIBJ), within the LAT-Teco2 project (National Rural Development Programme – PSRN), worked on developing a genetic evaluation for longevity in the Italian Jersey population. Cow stay-ability was analysed as a binary trait to estimate the ability of cows to survive until a specific calving event. The dataset included 20,724 cows raised to 1,135 herds and offspring of 544 sires, all showing at least a first calving event. The stay-ability curve (rate) was calculated and this helped to define the breeding objective, choosing survival at the fourth lactation. EBVs were estimated with a linear animal model, where the random effects included the herd-year of first calving, the animal and the residual error. The longevity index has been calculated as a composite index including information about animals at second, third and fourth lactations; weights were based on the population's stay-ability rate per lactation. Bulls' daughters' stay-ability rate (DSR) to the fourth lactation was calculated as a phenotypic indicator. Bulls were then categorised into three classes based on their index value: sires with index value greater than 105 had 44.58% DSR, while sires with index value lower than 95 showed 20.58% DSR. The results suggest that this trait exhibits genetic variability and that selection can be applied to improve it. It will officially release as a new longevity index for Italian Jersey population at April 2024.

Estimating time-dependent genetic parameters and SNP effects for longitudinal body weights in Holstein cattle

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Growth traits are directly linked to economic returns in beef cattle, and more recently in dairy cattle herds employing “beef-on-dairy” crossbreeding. Body weight (BW) is also important in dairy cattle breeding schemes due to its associations with feed efficiency, methane emission, and health. This study aimed to estimate the pattern of genetic parameters and SNP effects over time for BW in Holstein cattle. BW records of 2,173 animals were repeatedly collected once a month from birth to 12 months of age using an intelligent weighing scale. A random regression model with maternal effects was applied to estimate variance components. SNP solutions were obtained for random regression coefficients, which were used to estimate time-dependent effects based on weighted single-step genome-wide association. The top 1% of SNP that showed a high magnitude of SNP effect in at least one month were selected as relevant SNPs and clustered using a fuzzy c-means algorithm. Direct heritability for BW were moderate to high (0.24 to 0.62), with higher values at the beginning and end of the curve. Maternal heritability for BW gradually decreased from birth (0.39) to 60 d of age, stabilizing around 0.1 afterward. A total of 3,484 SNPs were selected as the relevant. Five clusters were formed according to the trajectory of SNP effects over time. The majority of relevant SNPs increased their effects with age and then stabilized, while about 21 percent of relevant SNPs displayed large effects only at birth. Our results provide evidence of differential sets of SNPs underlying the phenotypic expression of the BW with age, contributing to a better understanding of the genetic architecture of growth traits in dairy cattle.

Session 71

Poster 22

Study of LSR, proxy for robustness of fattening pig, in several environments and the link with tail biting

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The aim of this study was to evaluate genetic parameters of LSR, a proxy for robustness of fattening pigs (Lenoir et al. 2023) in 2 environments and 2 breeds and make the link with other traits associated with robustness such as tail biting. Between 77 and 138 days of age, for each individual, daily weight and daily feed intake were recorded. A total of 9864 Pietrain NN and 5757 Duroc were controlled in 2 farms. For those animals, around 138 days of age, tails were observed and registered as a binary trait: 0 no presence and 1 tail biting observed, independently of the damage. The overall average of occurrence for tail biting was 2.1% in Pietrain NN and 0.4% in Duroc. LSR trait, was calculated for each individual using the methodology of Lenoir et al (2023) and analysed within the breed and the farm of control. Heritabilities of LSR in Pietrain were respectively 0.06 (+/-0.03) and 0.071 (+/- 0.01) for farm1 and farm 2 separately and 0.07 (+/- 0.007) for all the Pietrain. In Duroc, heritability shifted from 0.04 (+/-0.01) to 0.07 (+/-0.01) depending which environment we considered. Standardised EBV for are on average 0.20 for animals with tail biting observations and 0 for animals without in Piétrain NN, 0.10 and 0 in Duroc. Heritability is quite stable depending of the quality of the data and it is significantly different from 0. It seems that selection for reducing LSR can have a favourable impact on the risk of tail biting. This has to be confirmed for other traits linked to robustness such as the ability to be selected at 100kg (R2 or R3, Lenoir et al 2021).

Genetic variation of HSP70.1 locus in Cinisara and Modicana cattle breeds

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Heat Shock Proteins (HSPs) are known to play crucial roles in environmental stress tolerance and thermal adaptation of animals. Among the HSP gene family, HSP70.1 is believed to be temperature sensitive and induced by physiological, pathological and environmental stressors. In a preliminary trial the polymorphism of HSP70.1 gene (also known HSPA1A) was investigated in two Sicilian native cattle breeds reared under hot climate conditions and, for comparison, in Italian Holstein (IH). A representative sample of 117 cattle (30 Cinisara, 30 Modicana and 57 IH) was analyzed for 4 SNPs loci (g.2154 G/A, g.64 G/T in 3'UTR and g.895 C/- and g.1128 G/T in 5'UTR). Haplotype frequencies were determined by fastPHASE. All the SNP markers were found to be polymorphic in the Sicilian breeds, except for 3'UTR g.2154A/G which was monomorphic for g.2154A as in IH. Largely predominant alleles were observed at each locus in the whole sample, with frequencies of up to 95%, 80% and 77% (at 3'UTR g.64G/T, 5'UTR g.895C/- and 5'UTR g.1128G/T, respectively) in the two Sicilian breeds. The 5'UTR g.895C/- locus was the most polymorphic, with three genotypes in all breeds. The deletion, associated with high expression of the HSP70 protein in response to heat stress, showed the highest frequency in Cinisara cattle. In both the Sicilian breeds, 5'UTR SNPs were found in linkage (r^2 0.86) and the 5 inferred haplotypes showed a rather homogeneous distribution. The observed genetic variation provides new insights into the genetic determinism of thermotolerance in these native breeds.

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Poster 24

Variance components and Genetic parameters for live weight in Dairy heifers in New Zealand

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Live weight (LW) is a significant trait in dairy and beef cattle production in New Zealand. Cattle originating in the dairy industry make up over 60% of the beef produced in New Zealand. In the future, genetic evaluations of dairy cattle may need to include additional indexes that can evaluate the beef worth of dairy females. The ideal dairy × beef calf has a low birth weight, to be born easily, but grows rapidly to slaughter weights. Dairy replacement females are routinely weighed, mostly between weaning and entry to the dairy herd, providing a large dataset for live weight and growth pattern evaluation. This study aimed to estimate variance components and genetic parameters for LW in the New Zealand dairy heifer population. A total of 2,564,927 LW records from birth to 24 months for 1,669,849 dairy heifers born between 2008 and 2022, along with a full pedigree of 4,864,842 animals, were analyzed using a multi-trait model. LW at different ages was defined: LW1 (birth to 1 month), LW2 (6 months), LW3 (12 months), LW4 (18 months), and LW5 (24 months). The analysis was conducted using APEX – Linear Model Suite with a multi-trait model. The heritability of LW decreased from 0.40 for LW1 to 0.35 for LW2 before gradually increasing to 0.53 for LW5. The genetic correlation was lowest between LW1 and LW5 (0.70), whereas the highest genetic correlation was found between LW4 and LW5 (0.98). These results provide valuable insights into the change in variance components and heritability estimates by age before maturity. Additionally, the high genetic correlation between LW3 and LW5 (0.92) suggests that LW at age of 12 months can serve as a proxy to predict breeding values for mature LW.

Single-step genome-wide association for lameness and BCS in Walloon Holstein dairy cows

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Lameness is recognized as one of the most important welfare challenges in dairy cows, causing pain and impairing the animal's ability to exhibit normal behavior. Lameness is associated with reduced milk yield, increased culling, and impaired reproductive performance. Furthermore, a low body condition score (BCS), possibly due to decreased thickness of the digital cushion, is a risk factor for lameness in dairy cows. This study aimed to perform genome-wide association (GWAS) studies to identify genomic regions associated with lameness and BCS in Walloon Holstein cows. Data were collected between 2018 and 2022 on 12,988 animals distributed in 307 herds in the Walloon region of Belgium. Lameness information is rated on a scale of 1-5: 1 = sound, 2 = mildly lame, 3 = moderately lame, 4 = lame, and 5 = severely lame. Animals were assigned a BCS score based on a nine-point unit scale with low values reflecting emaciation and high values corresponding to obesity. Data of 563,882 single nucleotide polymorphisms (SNP) located on 29 *Bos taurus* autosomes (BTA) on 4,628 animals (1,591 males) were used. The SNP solutions were estimated using a single-step genomic BLUP approach. The proportion of the total additive genetic variance explained by windows of 0.5 Mb was calculated, and regions accounting for at least 0.50% of the total additive genetic variance were identified. Mean (SD) lameness and BCS were 1.36 (0.66) and 5.40 (0.79) units, respectively. The average prevalence of lameness (defined as grades 2 to 5) was 27.53%. Heritability estimates for lameness and BCS were 0.02 and 0.25, respectively. The genetic correlation between BCS and lameness was -0.19, indicating that lower BCS is associated with an increased risk of lameness in dairy cattle. The GWAS results revealed that a region located from 78.7 to 79.2 Mb on BTA15 (harboring PTPRJ, OR4S1, and OR4X1 genes) is associated with lameness, and regions between 88.57 to 89.01 Mb in BTA6 and 71.23 to 71.73 Mb in BTA20 (harboring NPFFR2, CLPTM1L, TRIP13, TERT, CEP72, and NKD2 genes) were associated with BCS. The findings of this study provide a better understanding of the genomic architecture underlying lameness and BCS in dairy cows.

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Poster 26

Patterns of occurrence of mendelian errors in snp array genotypes in a src iberian pig population

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The ascertainment of the reliability of the genotypes obtained using SNP arrays is a major issue in livestock genetics. Calling errors lead to editing genotypic data (usually considering minor allele frequency or Hardy-Weinberg proportions) causing major losses of information. The aim of this study was to ascertain whether family structure and genomic features could underlie Mendelian Errors (ME) from a selection experiment to decrease environmental variance for birth weight in an Iberian pig population (Sánchez Romero Carvajal). These were identified in 38 parents-offspring trios of Iberian pig genotyped using the Axiom-PigHDv1 array (658,692 SNPs retained). ME were classified into 8 classes defined according to the trio member to which the ME was assigned (Trio, Father, Mother or Offspring), being the Trio class when the ME could not be assigned to an specific member of the parent-offspring trio, and the allele on which the ME was identified (A: major allele, B: minor allele): TrioA, TrioB, FatherA, FatherB, MotherA, MotherB, OffspringA and OffspringB. A total of 18,326 mismatches were identified on 8,966 different loci. Most mismatches were assigned to both the Father (39.5%) and the Mother (39.8%). Between 36.4% and 39.7% of the loci with ME assigned to the parents had Allele-Drop-out events. The ME class involving a higher number of loci was TrioB (2,693 SNPs), 55.8% of which had Allele-Drop-in events. The distribution of ME classes is consistent with previous reports in pig population.

GWAS for productive traits in Italian Alpine and Saanen goats

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Recently, genomic estimated breeding values have been developed in the Italian Alpine and Saanen goats for productive traits related to milk yield and quality. In this work, we applied a single-step genome-wide analysis (GWAS) to identify positional and functional candidate genes related to these traits. The analyses included 65K SNP data, 210-d lactation records, and pedigree information for 2472 Alpine and 1139 Saanen goats. The GWAS was performed using blupf90+ and postGSf90 software. We identified 245 and 276 significant SNPs in Alpine and Saanen breeds, respectively. In both breeds, we found a peak on the region of CHR6 harboring casein genes (79-87 Mb) associated with protein percentage and two peaks on CHR6 (84-86 Mb) and 14 (84-85 Mb) associated with fat percentage. These results are consistent with similar studies in goats; moreover, these regions are known to regulate milk quantitative traits in cattle. Furthermore, significant SNPs for milk yield in Saanen goats only presented a peak on CHR19 (25-28 Mb). Increasing our knowledge of the genomic regions associated with improved milk yield and quality could enhance the selection of the Italian Alpine and Saanen breeds.

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Poster 28

Exploring the genomic basis of shank and eggshell coloration in Italian native chicken breeds

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Eggshell and shank colour play a role in poultry selection, breed recognition, and environmental adaptation. This study investigated the genomics of shank and eggshell pigmentation in Italian local chickens to uncover the mechanisms underpinning these phenotypes. To this purpose, 418 animals from 18 local breeds were evaluated for shank and eggshell pigmentation, and genotyped using the Affymetrix Axiom 600 K Chicken Genotyping Array. A genome-wide association study was performed to compare the case and control populations for shank and eggshell colour. The most interesting locus for shank pigmentation was detected on GGAZ, close to the TYRP1 gene, which plays a key role in avian pigmentation. Also, novel loci and genes (e.g., MTAP, CDKN2A, CDKN2B) associated with shank and skin pigmentation, UV protection, and melanocyte regulation were identified. A new genomic region for shank pigmentation was detected on GGA1, in which CHODL, TMPRSS15, and NCAM2 gene were mapped. Fewer significant loci were identified for eggshell pigmentation, including SLC7A11 on GGA4 and MITF on GGA12, which are associated with melanocyte processes and pigment synthesis. This study shed light on the genomic architecture underlying shank and eggshell colour in Italian local chicken breeds. These phenotypes play an important role in breed identification and conservation. This work was funded by MIPAAFT (project TuBAvI, PSRN 2014-2020, sottomisura 10.2).

RESILTROUT investigates the trout genome to identify marker associated to resilience to diseases and heat stress

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The RESILTROUT project aims to enhance Italian trout farming competitiveness through research and technology integration. Specific objectives include achieving environmentally friendly businesses, selecting resilient trout strains for climate adaptation, implementing water recirculation systems, promoting circular economy practices, reducing pharmaceutical contamination, protect biodiversity, and ensuring fish welfare. Case-control Genome-Wide Association Studies (GWAS) are conducted to investigate traits associated to trout farming economic and environmental sustainability. Two studies focus on identifying genetic markers associated with resistance to two diseases, lactococcosis and Proliferative kidney disease (PKD). This will aid in breeding programs to develop trout strains with enhanced resistance, reducing the reliance on antibiotics and promoting environmental sustainability. Another GWAS targets heat stress resistance, crucial for reducing mortality rates during temperature fluctuations. By comparing genetic variations between cases (dead fish) and controls (surviving fish) under heat stress and/or natural disease challenge conditions, key genetic factors contributing to resilience can be identified. Implementing findings from these studies will enhance the resilience and health of trout populations, contributing to the overall sustainability and competitiveness of Italian aquaculture.

Culling reasons for end of career dairy cows in northern Italy

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Cows may be at the end of their career, and therefore culled, because they are no longer productive but still fertile and healthy (voluntary culling), or because they are lame, injured or sick (involuntary culling). In this study, 7 dairy farms were visited in 2022 to analyse cow culling rates and reasons for culling in northern Italy. Data were related to adult cows (parity \geq 1) and obtained from the herd management software and through discussions with the farmers. Information collected for each culled cow included parity order, days in milk (DIM), type of exit from the herd (i.e., sale, slaughter, found dead, emergency slaughter (OFES)), and main reason for culling. Results showed variable herd size (average lactating cows=236; min=59, max=586) and milk yield (average 37 kg/day/head; min=32; max=39.7). Out of a total of 1,898 Holstein Friesian adult cows exposed to the risk of culling, 869 (45,8%) were actually culled: 36 (4,1%) for OFES; 54 (6,2%) found dead; 365 (42,0%) sold to another farm; 414 (47,6%) sold for slaughter. Median lactation at culling was 2 (min=1; Q1=1; Q3=3; max=9); median DIM was 126 (min=1; Q1=51; Q3=243; max=1011). The reasons for culling were poor reproductive performance (26.1%), low milk yield (15.8%), high somatic cell count/mastitis (14.7%), unknown reason (12.3%), traumatic injuries (6.0%), udder defects/poor body conformation (5.8%), metabolic/digestive disorders (5.4%), premium prices (4.8%), old age (4.7%), other health issues (4.4%). Health issues and the early age at culling should raise questions about cow welfare, economic sustainability and environmental impact of dairy farming, suggesting areas for improvement to reduce involuntary culling and support a longer, high-quality productive life for cows.

Prioritizing markers for stayability in beef cattle and its impact on genomic predictions

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Stayability is the ability of a cow to remain in the herd, given it produced a certain number of calves. This trait is important as it relates to the longevity and profitability of the cow herd. We aimed to investigate the prediction accuracy of EBV for two definitions of stayability (STAY) using SNP-prioritized panels. A total of 200,180 Nel-lore cows were evaluated for two binary STAY traits: STAY63 and STAY64, defined as cows succeeding or not in calving 3 or 4 times until six years of age, respectively. Pedigree was available for 2,042,151 animals, from which 11,176 sires were genotyped for a 300K panel. Pedigree-based EBV were estimated, and genotyped sires were divided into high and low-performance groups based on the deviation of their EBV from the population mean. Standard deviation (SD) thresholds of 1, 1.5, and 2 were used. Sires in the high and low-performance groups had their genotypes compared with Wright's fixation index (FST), and SNP panels with the 10K, 25K, 100K, and 150K SNP with the highest FST scores were created. Genomic EBV (GEBV) were estimated with the single-step method with FST panels in the three SD scenarios. Prediction accuracy was calculated for a validation set of 61 genotyped bulls with the LR method. Results from panels of the same density composed of randomly selected SNP were used as the benchmark. Panels consisting of 10K and 25K selected SNP using the threshold of 2 SD presented the highest prediction accuracy among all scenarios and traits. The relative increase in accuracy ranged from 9 to 10.2% and from 7.7 to 8.2% for 10K and 25K panels, respectively. Selecting SNP based on FST scores enabled the use of lower SNP panel density while increasing prediction accuracy.

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Poster 32

Genomic study of milk performance in fat-tailed and thin-tailed Mediterranean dairy sheep

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The objective was to compare the genetic profiles of lactation milk yield (LMY) and fat (FC), protein (PC), and lactose content (LC) between the fat-tailed Chios and thin-tailed Frizarta sheep. We employed phenotypic and genomic data to: (i) derive heritability estimates and (ii) identify single nucleotide polymorphisms (SNPs) and candidate genes associated with the studied traits. A total of 317 Chios and 346 Frizarta ewes from four farms were included. Individual records of daily milk yield, FC, PC, and LC were collected monthly for two years and used to calculate LMY and weighted means of FC, PC, and LC. Genotypes were obtained with the Illumina OvineSNP50 Genotyping Beadchip. For each breed, variance components of studied traits were calculated with ASReml, and Genome-Wide Association Studies were performed with GEMMA software. Studied traits were equally heritable in both breeds (Chios: $h^2=0.42-0.70$; Frizarta: $h^2=0.34-0.62$) and followed a polygenic inheritance pattern. One genome-wide and 11 suggestive significant SNPs, and 28 candidate genes were associated with Chios sheep performance. Seven suggestive SNPs and 15 candidate genes were detected in the Frizarta data. No common SNPs or genes were identified between breeds. Results suggest a distinct genetic control of milk performance in the two breeds, possibly related to the diverse evolutionary trajectories of fat-tailed and thin-tailed sheep. This work was funded by the SMARTER (772787) and iSAGE (679302) Horizon 2020 projects.

Preliminary investigation of the reaction norm model for the milk yield in Taiwan's Dairy Herd Improvement data

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Taiwan is under the tropical/subtropical oceanic climate with high temperature and humidity for about 8 months in a year. Genotype by environment interactions arise when the performance diverse among two or more genotypes varies across tropical/subtropical or temperate environments. Consequently, the objective of this preliminary study was to obtain the genetic parameters from the reaction norm model to investigate the genotype by environment interactions in Taiwan dairy herds. The data and pedigree from the Dairy Herd Improvement program (DHI) in Taiwan were collected during 2009 ~ 2022. In total, 101,677 lactation records from 55,673 cows were obtained from 126 herds and there were 703,356 animals in the pedigree. Two models were applied for the milk yield, including the conventional animal model (AM) and the reaction norm model (RNM). Both models include herd/year/season, lactation and physical situation of lactation as fixed effects, then additive genetic effect and permanent environmental effect as random effects. The main difference between the two models is that the year-season solutions (YSS) obtained from AM were added as a covariate to obtain the slope of the animal additive genetic effect in RNM. The result showed that the heritability under AM was 0.219 and constant throughout YSS. However, the heritability under RNM showed genetic variability under different YSS ranging from 0.156 to 0.294, which increased with better environment conditions. The genetic correlation between YSS ranging from 0.908 to 0.999, which decreased gradually as the difference between environmental gradients increased. This preliminary study showed the presence of genotype by environment interactions in Taiwan's dairy herds might exist, and further study on the reranking of dairy cows/sires in production traits is needed to be investigated.

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Theatre 1

Including QTL information in GBLUP and machine learning models for genomic prediction

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With the accumulation of genotyped individuals, increasingly more quantitative trait loci (QTL) are being detected. Including QTL genotypes in prediction models can potentially improve the accuracy of predicted breeding values. It is expected that machine learning (ML) models would benefit from including QTL genotypes in the prediction model, because ML models can put weights on variables intrinsically. The aim of this study is to determine the benefit of including QTL information in ML models for accuracy of genomic prediction. We simulated datasets of populations under selection and the proportion of genetic variance explained by the preselected QTL was varied from 0 to 100%. Two ML models, random forest (RF) and support vector regression (SVR), were used. In the ML models, QTL were included among the SNPs dataset for genomic prediction without giving extra weight. We compared the accuracy from ML models to a 2GBLUP model in which different proportions of QTL genotypes were included as a separate genetic effects. The results showed that prediction accuracy of the 2GBLUP model was slightly higher than SVR and much higher than RF for all scenarios, and that RF resulted in the lowest prediction accuracy. For 2GBLUP, prediction accuracy increased approximately linearly as long as <80% of the total genetic variance was explained by the QTL, while accuracy was reduced when more QTL were included. For SVR, accuracy continuously increased with more QTL included in the model, while the increase was smaller than that of 2GBLUP. For RF, however, accuracy did not increase. In conclusion, the benefit of including QTL genotypes depended on the used ML and was lower for SVR and RF than for 2GBLUP.

A Nonlinear Recursive Model for Predicting Calving Difficulty and Birth Weight in Cattle

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Calving difficulty (CD) in beef cattle poses significant challenges, leading to increased calf losses, cow mortality, and heightened veterinary and labor costs. It is influenced not only by the dam itself but also by its calf, categorized as maternal and direct effects, respectively. Traditionally, calving difficulty has been attributed to calf size or weight. In this study, we employed recursive models to dissect the influence of the calf and its weight on calving difficulty. Our dataset comprised 264,627 records of calving difficulty and birth weight, with calving difficulty measured on a four-category scale (1. No help – 68.59%, 2. Moderate Help – 26.91%, Strong Help – 3.39%, and 4. Caesarean – 1.11%). The average birth weight was 41.58 kg, with a standard deviation of 6.58 kg. The pedigree encompassed 522,885 individual-sire-dam entries and we dispose of 4,578 individuals genotyped with the Axiom™ Bovine Genotyping v3 Array. For the analysis, we utilized a threshold-linear bivariate model under a single step-approach, incorporating factors such as sex, dam age, year-season, herd, permanent maternal, genetic maternal, and direct effects. Our findings revealed that the direct effect of the calf explained a larger percentage of the total variance than the maternal genetic effects, even after implementing recursive models with birth weight. We explored both linear and non-linear recursive models, comparing them using the Deviance Information Criterion (DIC). The non-linear model exhibited the best fit, suggesting a non-linear relationship between traits in both the liability and observed scale. This non-linearity implied that the heritability of calving difficulty and the genetic correlation with birth weight evolve across the parametric space of birth weight. Finally, we calculated the SNP effects and the amount of genetic variance associated with each genomic region by backsolving from the prediction of breeding values to illustrate how the distribution of the genetic variability of calving difficulty is modified with the calf birth weight.

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Theatre 3

Effect of genotyping density on the detection of runs of homozygosity and heterozygosity

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Runs of homozygosity (ROHom) are continuous stretches of homozygous loci that can reflect autozygosity, whereas runs of heterozygosity (ROHet) are heterozygosity-rich regions that can contain genes resilient to genetic drift. The present study aimed to test the effect of chip density in detecting ROHom and ROHet in the Italian Simmental cattle breed. A sample of 897 animals were genotyped at low- (397 individuals), medium- (348 individuals), or high-density (152 individuals) array. The number of ROHom and ROHet per animal and their average length were calculated. Runs shared by more than one animal and the number of times a particular SNP was inside a run were also computed. The ROHom number increased, their average length decreased, and the distribution across length classes varied as the chip density increased. On the contrary, the ROHet number decreased, and the average length increased as the chip density increased; no ROHet were found using the high-density chip. Thus, the results of the present study confirm that the detection of ROHom is more reliable when the chip density increases, whereas the ROHet trend seems to be the opposite. Genes and QTL mapped in the regions highlighted by both approaches confirm that ROHet can be due to balancing selection and, thus, related to fitness, health, and reproduction traits. In contrast, ROHom are mainly involved in production traits.

Exploiting properties of the H-Matrix as an aid to identify Metafounders

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The Metafounder (MF) approach was developed to bridge the conceptual gap between the concept of identity by descent underlying the numerator relationship matrix A and the genomic relationship matrix G . In a recent publication we investigated the feasibility of defining MF based solely on genotype data. We used population structure analysis of genotyped (gt) animals to identify stratifications in the population and to assign MF. Such a characterisation of MF improved the compatibility of A and G for gt animals. However, there is no straightforward way to extrapolate this assignment to the pedigree founders (PF), which are in most cases not genotyped. The matrix of relationships between MF (Γ) establishes an approximation of the existing relationships/stratifications between PF, which are unobservable from both A and G . We argue that this is in some sense similar to aspects of matrix H , the common additive-genetic covariance between non-gt and gt animals used in Single-Step. In H , approximate relationships between non-gt animals are established based on the observed genotypes of related animals. We exploited this similarity by using a clustering algorithm on HPF, PF , the submatrix of PF , to assign PF to MF. To illustrate the approach, we clustered PF for a predefined number of MF, estimated MF allele frequencies, set up Γ and constructed $A\Gamma$ for a sample from the Fleckvieh population spanning multiple generations of genotyped animals. Based on these results we show consequences of MF use in breeding value estimation for a modern dairy cattle population.

Session 72

Theatre 5

The effective number and profile of causative markers in GWAS

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Over the past decade, animal databases have amassed abundant genomic data, with gradual progression from using thousands to using millions of markers. Against expectations, this increase in marker density only marginally increased the predictivity of the standard quantitative genetic models. To reason for this, we used stochastic simulations and statistical modelling to understand the marker profile around the quantitative trait nucleotides (QTN) in populations with a small effective population size (N_e). We applied the single-step genome-wide association to simulated datasets with varying N_e and number of genotyped individuals with phenotypes. We decomposed resulting Manhattan plots into signals from the QTN itself, QTN profile, relationships, and noise. In this sense, the QTN profiles can be understood as pairwise linkage disequilibrium curves, with width inversely related to N_e . Thus, in larger populations, QTN profiles were narrow, which, coupled with weaker relationships, facilitated QTN identification (with ample phenotypic data). Conversely, smaller populations yielded wider QTN profiles that blended with relationship signals and noise, resulting in reduced GWAS resolution and QTN identification. In terms of predictivity, predictions in large populations required dense marker panels and QTN or nearby markers identified. In contrast, in smaller populations, medium marker density sufficed as the QTN profiles captured the QTN without its explicit identification.

Modeling unknown parents in single-step genomic BLUP: Metafounders vs. unknown parent groups in a simulated cattle population

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The aims of this study are to compare of methodologies involving unknown parent groups (UPG) and metafounders (MF), to propose and validate a method for categorizing MF based on genotypic data, and to assess various approaches for estimating the Gamma matrix across different scenarios within a simulated dairy cattle population characterized by varying degrees of pedigree completeness. In scenarios where unknown ancestors are rare, the impact of UPG or MF on breeding values is minimal, however, MF demonstrates slight enhancements over UPG. Conversely, scenarios characterized by lower genotyping rates and higher occurrences of unknown parents reveal notable differences in evaluations with and without UPG, as well as in comparison to MF. Specifically, incorporating UPG via Quaas-Pollak-transformation in the pedigree-based and genomic relationship matrix using ssGBLUP (UPG_fullQP) results in noticeable bias and overdispersion. This study emphasizes the significance of incorporating crossbreed genotypes in the estimation of the Gamma matrix for improved accuracy. Categorizing UPG or MF is a challenge, nevertheless, the genotype-based method used in this study provides predictions comparable to true group assignments. Validation results obtained using the linear regression method indicate a slight superiority of MF evaluations, but with much smaller magnitudes compared to true validations. Remarkably, the extreme bias observed in UPG_fullQP is not detected using routine validation statistics.

Confidence intervals for validation statistics with data truncation in genomic prediction

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Validation by data truncation is a common practice in genetic evaluations because of the interest in predicting the genetic merit of a set of young selection candidates. Two of the most used validation methods in genetic evaluations are predictivity (correlation between pre-adjusted phenotypes and EBV) divided by the square root of the heritability) and the linear regression (LR) method. Both methods compare predictions with the whole and partial datasets obtained by removing the information related to a set of validation individuals. Confidence intervals (CI) for predictivity and the LR method can be obtained by k-fold validation or bootstrapping. Analytical or frequentist CI are unavailable for predictivity and the LR method and would be beneficial to avoid running several validations. The analytical CI can also help test the quality of bootstrap intervals. This study aimed to derive analytical CI for predictivity and statistics included in the LR method (bias, dispersion, ratio of accuracies, and reliability). The CI for the bias, dispersion, and reliability depend on the (co)variances of the EBV across the individuals in the validation set. The CI for the ratio of accuracies and predictivity were obtained through the Fisher transformation. We showed the adequacy of the analytical CI using simulation. The analytical CI were closer to the simulated ones. Bootstrap CI tend to be narrower than the simulated ones. Estimating the sampling variation of predictivity and the statistics in the LR method without replication or bootstrap is possible for any dataset with the method proposed in this study.

Multibreed single-step GBLUP evaluations for fertility traits in US dairy cattle

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We tested ssGBLUP for fertility traits mimicking the CDCB Dec-2022 official run. We included all phenotypes for Daughter Pregnancy, Cow Conception, and Heifer Conception Rates, and Early First Pregnancy, comprising 94 million records. The pedigree contained 94 million individuals across six breeds and their crosses and 417 Unknown Parent Groups (UPG) modeled as unrelated or metafounders. We used about 2 million genotyped animals in ssGBLUP with the algorithm for proven and young. A total of 45K genotyped animals representing the breeds and crosses were set as core. Results from the ssGBLUP run were compared with those from BLUP regarding dispersion bias and prediction reliability. The whole dataset contained phenotypes up to Dec-2022, and the partial dataset was truncated in Dec-2018. We compared (G)EBV of bulls with no daughters with records in the partial and at least 100 daughters with DPR records in the whole dataset, resulting in 1891 bulls for Holstein and 303 for Jersey. The linear regression method was used for validation. The regression coefficients varied between 0.89 and 0.94 for ssGBLUP with metafounders and 0.88-0.90 for UPG, resulting in a slightly biased evaluation. For BLUP, the values were from 0.76 to 0.93 and 0.72. to 0.92, respectively, which is more biased because BLUP cannot account for the genomic selection of young animals. The correlation between early and late proofs from BLUP varied from 0.49 to 0.65 for metafounders and 0.50 to 0.63 for UPG. For genomic proofs, they ranged from 0.80 and 0.91 with metafounders and 0.81 to 0.89 with UPG, showing good reliability of early genomic proofs. Overall, ssGBLUP seems viable for the genomic evaluation of fertility traits in US dairy cattle.

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Theatre 9

Comparative transcriptomic profiling of mRNA isoforms linked to feed efficiency in Holstein dairy cattle

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Genomic and nutritional strategies to improve cattle feed efficiency (FE) may lead to environmental and economic benefits for the dairy industry. Transcriptomics using RNA-Sequencing (RNA-Seq) can detect differentially expressed (DE) mRNA isoforms that may impact metabolic efficiency in cattle. Our study identified novel and annotated mRNA isoforms DE between low and high FE [Residual Feed Intake (RFI; kg/d)] Holstein cattle which were fed two diet treatments. RNA-Seq data from liver samples of high-RFI (n=4) and low-RFI (n=5) Holstein cows fed a control (C; n=9) and high concentrate (HC; n=9) diet were used. Using CLC Genomics Workbench, significantly DE novel and annotated mRNA isoforms (FDR<0.05; |FC|>2) were identified between RFI groups within each diet, and between diet groups within RFI using Large Gap Read Mapping. Comparisons between low- and high-RFI groups revealed 31 and 21 DE mRNA isoforms in low- vs. high-RFI with the C diet and low- vs. high-RFI with the HC diet respectively. Among them, TDG_2 and BRIP1_7 were the most significantly DE mRNA isoforms and are involved in DNA repair, gene expression, metabolism of proteins and cell cycle processes. In addition, 19 novel DE mRNA isoforms were identified including Gene_6794_4. Intra-group comparisons within low- and high-RFI animals revealed 25 and 18 DE mRNA isoforms between C and HC diets in low- and high-RFI respectively. Metabolic pathway analysis was performed by Reactome using the lists of DE mRNA isoforms. A total of 214 pathways were significantly enriched (FDR<0.05) between RFI groups (155) and between diets (59) including amino acid metabolism and myogenesis. Further analyses will focus on the detection of potential SNPs causing those alternative splicing events. With enhanced genetic insights into the regulation of FE this could optimize selection in the dairy cattle industry.

Exploring modelling approaches to address the dynamic nature of animal health

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Health is usually considered in a binary manner: the animal is healthy or not, for a given disease or disorder, at a given time. However, health is a multifactorial status and evolves through time. The development of precision livestock farming and high-frequency phenotyping provides trajectories of performances or traits that can be used as indicators of the health status, and allow its dynamic nature to be taken into account. In this study, we explored conceptual approaches to model the evolution of the health status of an animal facing a disturbance. The animal's observable reaction through time (or degree of health) was considered as a function combining the intensity of the environmental disturbance and the biological ability of the animal to cope with the perturbation, the latter being a function as well, depending on individual factors such as genetics, current physiological state or life course, for example. Using a sigmoid relation between the degree of health and the intensity of disturbance seems a promising option. It implies that low levels of intensity do not affect health, and that there is a maximum degradation of health for high levels of intensity. We expect that having dynamic trajectories of a high number of animals subjected to the same environmental disturbance intensity would enable the characterization of individual resilience, as well as estimating the unmeasured dynamic trajectory of the environment intensity. This is currently being tested on real data.

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Poster 11

The impact of truncating historical data on the prediction ability of selection candidates

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Breeding programs implemented routine genetic evaluations several decades ago. Since then, the development of molecular techniques, bioinformatics and evaluation methodologies have revolutionized the estimation of breeding values, but pedigree and phenotypic records are still on the heart of the evaluation. At this point, an important question in genetic evaluations is whether considering old generations of pedigree, and phenotypes (even genotypes) is beneficial or a disadvantage. Thus, the purpose of this study was to evaluate the impact of removing historical data combined with two pedigree deepness and the inclusion of genomic information, in the prediction ability of genotyped young Latxa dairy sheep individuals. 120 days standardized milk yield records of Latxa Cara Negra from Euskadi population were used at six different deletion scenarios, from 1992 to 2012 every four years, which were assessed by cross-validation with the LR method, and bias and accuracy statistics were calculated. The prediction accuracy of selection candidates when genomic information was included was the highest (0.69), and deletion of historical phenotypic data had the biggest gain of 1.5 % when data before 2000 was removed. The bias of validation individuals was lower when the EBV was predicted based on genomic information (-2.1), being lower when the biggest amount of data was deleted. Thus, removing old data until 2000 or 2004 resulted in reduced prediction bias, without compromising or even increasing prediction accuracy of selection candidates. The obtained results for ssGBLUP evaluations were outstanding, compared with traditional BLUP evaluations, while depth of pedigree had a very small influence.

Genetic diversity of German and Irish Beef cattle breeds

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In 2023, single-step genomic evaluation was introduced into the national breeding evaluation of 6 German beef cattle breeds. Genetic diversity is essential in ecological, genetic, and political discourse today. Managing genetic diversity in livestock populations is vital to sustainable livestock production. This study presents a detailed assessment of genetic diversity in five beef cattle populations based in Germany and Ireland. We analysed 186,509 individuals using 37,294 autosomal SNPs. We analysed the genetic structure and characteristics at the breed, population, and national levels. The higher levels of inbreeding in the Irish beef cattle breeds indicate a higher intensity of breeding in the Irish beef cattle industry. The PCA analysis showed a tighter cluster in the German beef cattle breeds, suggesting that German beef cattle breeders source their cattle from a similar genetic pool. The results from this study show the close genetic relationship between German and Irish beef cattle populations. This study supports biochemical and molecular genetic studies, which reflect breeds with a common geographic origin or single breeds that have expanded by export and crossbreeding. Our result is essential in the management of cattle genetic diversity as well as the design of international breeding and population improvement programs in beef cattle, particularly between Germany and Ireland. As the global agricultural landscape continues to evolve, these findings provide a robust foundation for informed decision-making and strategic planning in the realm of cattle breeding and population management.

An evaluation of using Runs of Homozygosity to detect Identity by Descent

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Identity by Descent (IBD) serves as a vital tool for gene mapping, relatedness estimation, and demographic history exploration. However, its measurement poses challenges, leading to the use of Runs of Homozygosity (ROH) as an alternative metric for IBD. However few studies have examined the accuracy of using ROH as a proxy for IBD in livestock populations. Therefore, this study aims to determine the extent to which ROH are IBD and identify portion of IBD that goes undetected. We simulated 100 generations of randomly mated animals with an additional set of markers tracing IBD. ROH were detected using 4 different settings. Using the founder markers, we calculate the true positive rate (TP), false negative rate (FN), and correlation between FROH and FIBD. Additionally, within-ROH inbreeding (F|ROH) was calculated by comparing the increase in IBD compared to the general expectation in the genome. Only ROH over 9Mb were consistently 90% IBD, regardless of distance to base population. A high marker density was essential to achieve high TP and low FN. Despite of the generally low TP, the F|ROH was over 70% for all bins of ROH and parameter combination. As an individual measure of inbreeding having a high marker density and higher minimum ROH length threshold was important to achieve correlations over 0.9 and regression coefficients close to 1 when comparing FROH and FIBD. For a 150k and 300k SNP chip ROH is only an adequate and stable indicator for IBD if the ROH are longer than 9 Mb. Thus, for projects interested in tracing ancient inbreeding, a significantly higher SNP density is required.

Screening continent-wide genomic diversity for local adaptation reveals strong signals of spatially divergent selection in European sheep

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After livestock domestication and dispersal, environment-mediated selective pressure has shaped phenotypic variation and left specific signatures in the genome of locally adapted breeds. The identification of genes of adaptive relevance is strategic for the livestock sector, especially to prioritise genomic resources for conservation and to guide selection in a period of rapid climate change. Among domesticated species, sheep (*Ovis aries*) have established a wide geographic range due to their rusticity, manageable size, adaptability to nutrient poor diets, and tolerance to highly diverse environments and climatic conditions. Within the framework of the IMAGE project (H2020-SFS-2015-2/677353), we used high-density SNP genotypes from 80 autochthonous sheep breeds, spanning from North Africa to Scandinavia to sample a wide range of climatic conditions. We investigated the structuring of genetic diversity, and combined uni- and multivariate gene-environment association approaches (LFMM and pRDA) to pinpoint genomic regions candidate for local adaptation to diverging climates. Results pointed to energy management, water retention and DNA reorganisation as relevant for climate adaptation. Finally, we computed a local genomic offset metric and identified moderate to severe adaptation challenges in most of the European sheep breeds.

Genetic markers to differentiate domestic pigs and wild boars

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Identifying species in food and feed products is important in human and animal diets, as well as preventing the spread of various diseases (e.g., ASF). It is also relevant in forensic investigations involving animals (e.g., bites) or potentially adulterating food products. An efficient and cost-effective method has not yet been developed to distinguish between domestic pigs (*Sus scrofa domestica*) and wild boars (*Sus scrofa scrofa*). The study aimed to analyse the polymorphism of the MC1R and NR6A1 genes and evaluate their usefulness in distinguishing between both subspecies. DNA extracted from hair follicles and skin samples, and peripheral blood from 22 wild boars and 53 pigs of five breeds (Polish Large White (PLW), n = 14; Polish Landrace (PL), n = 13; Puławska (P), n = 10; Duroc (D), n = 17) was examined. Two samples representing hybrids created by mixing DNA from two pig and wild boar samples were analysed. Polymorphism of MC1R at g.370G>A, g.491C>T, g.727G>A was studied using PCR-RFLP. Identification of the C > T SNP at locus g.748 NR6A1 (GeneBank accession number AB248749) was performed using fluorescently labelled TaqMan MGB probes (VIC-TCACcGGGCTCCA-MGB NFQ FAM-CTCACtGGGCTCC-MGB NFQ) using a StepOne Plus device (Applied Biosystems, USA). Seven alleles and seven genotypes were identified in the MC1R gene. Additionally, three hybrids were detected. In the case of the NR6A1 gene, all analysed pigs were identified as genotype TT and all analysed wild boars were identified as genotype CC. No pig-wild boar hybrids were detected in the samples studied.

The usefulness of a panel of STR markers recommended for pig identification for pork traceability

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DNA profiling is significant for animal identification, and parentage verification and also increasingly important for meat traceability. Consumers frequently choose pork as their meat, so maintaining high standards of pork production, not just for commercial pigs but also for native pig breeds, is crucial. The purpose of this study was to determine the population structure of the native Puławska Pig (PUL, n=85), and 3 commercial pig breeds: Polish Landrace (PL, n=85), Polish Large White (PLW, n=74) and Duroc (DUR, n=84) with using 14 STRs recommended by ISAG for pig parentage verification. The PCR reaction was performed by the Type-it Microsatellite PCR Kit (Qiagen). The PCR products were analyzed using an ABI 3500xl sequencer. The obtained results showed genetic differentiation among breeds accounted for 18% of total genetic variability (AMOVA). Bayesian structure analysis (STRUCTURE) indicated that the four distinct genetic clusters obtained correspond to the four breeds studied. The genetic Reynolds distances (Θ_w) showed a close relationship between PL and PLW breeds and more distant relationship between DUR and PUL pigs. The genetic differentiation values (FST) were lower between PL and PLW and higher between PUL and DUR. The principal coordinates analysis (PCoA) supported classifying the populations into 4 clusters. Our study demonstrates that a panel of STR markers recommended by ISAG may also be useful for pig breed prediction and, in the future, for meat traceability which is especially important for the population of native or local pigs, such as the Puławska breed, which can provide meat that is high in quality.

Species identification of cervid using common pair of primers

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Interest in species identification research is mainly related to the need to monitor food products resulting from health issues, religious beliefs, consumers and economic reasons. Another common aspect of research is the analysis of microtrace, pseudomedical products or samples not intended for consumption, most often associated with forensic analyses. The object of research is often the tissues of wild animals such as cervid. The study aimed to develop a method for distinguishing species of closely genetically related animals, such as red deer, roe deer, Dybowski's deer, fallow deer, and moose. Polymerase chain reaction (PCR) based on oligonucleotide primers targeting the mitochondrial 12S rRNA gene was applied to the species identification. After comparing mitochondrial sequences of species analysed, primers were chosen, which allowed receive PCR product about length 459 bp (F:5'-CATACTGGAAAGTGTGCTTGGA; R:5'-TGGACAACCAGCTATCACCA). The use of common primers, specific for all tested species, will enable amplification regardless of the specific cervid species tested. The next step of analysis will be Sanger's sequencing, after that it can be differentiated into individual species. This way of analysis will allow quick identification of several species at the same time, but it will enable exact differentiation too in the case this necessity. The chosen fragment is very homologist, its similarity for all identified species is over 92 %.

Detection of heterozygosity-rich regions in sheepdogs and cattedogs

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This study explores the distribution of heterozygosity-rich regions (HRR) and HRR islands (HRRi) in eight dog breeds belonging to the FCI category Sheepdogs and Cattedogs. We investigated whether HRRi arises as a consequence of balancing selection and if HRRi occur in the same genomic regions in breeds with similar historical purposes. After quality control, the dataset included 160 animals and 143,709 autosomal SNP markers. Consecutive SNP-based approach showed in metapopulation overall 10,618 HRR non-uniformly distributed in the genome. Differences in the number and average length of HRR between breeds corresponded to the level of the observed heterozygosity within each of them. A total of 64 HRRi defined based on the frequency of SNP in HRR (top 0.1%) contained 1197 unique SNPs. Overlap of HRRi was observed among two or three breeds across chromosomes 1, 5, 11, 13, 14, 16, 18, 20, 22, 25, and 33 (9 times in 2 breeds and 4 times in 3 breeds). Inside all detected HRRi, 211 protein-coding genes were identified. These genes were annotated with many different processes, but mainly with cellular membranes, sensor organ development, neurodegenerative disease, cancer and metabolism of zinc and ATP. The effect of balancing selection on the genome of analysed breeds was tested by Tajima's D statistics. Positive D values detected in HRRi regions suggested that these regions could be affected by balancing selection. We hope our research will raise awareness about HRR and HRRi in dog populations and draw attention to the possible link between HRRi and balancing selection. This research was supported by grants APVV-17-0060 and APVV-20-0161.

Session 72

Poster 19

Using genomic and transcriptomic information for complex trait prediction

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Prediction of complex traits is a central aspect of quantitative genetics. Since the use of SNPs has improved breeding value estimation, new low-cost, high-output technologies are being sought to further optimize breeding value estimation. One option could be the use of gene sequence data from expressed RNA. Our study used a data set of a Japanese quail F2-cross phenotyped for phosphorus efficiency and related efficiency traits. The birds were genotyped, and ileum miRNA and mRNA gene expression were classified. The data set contained 481 quail with 4k SNP genotypes, 77 miRNA and 80 mRNA gene transcript abundances, and trait records. We modeled several best linear unbiased prediction (BLUP) models using the genomic information of the animals (GBLUP), the transcriptomic information of the animals (TBLUP), and models containing both information in one model (GTBLUP). However, since the assumption of independent random effects may be violated in GTBLUP, we additionally considered a model with interaction effects (GTIBLUP) and a model that removes the redundant information between random effects (GTCBLUP). We found that the addition of transcriptomic information can explain a higher proportion of the phenotypic variance of a trait and the accuracies of trait predictions are higher than those of the GBLUP model. In addition, transcriptome-wide association analyses identified large trait-specific effects of individual gene transcripts. Our results suggest that transcriptomic data is a promising additional data source to be used in animal breeding for explaining phenotypes and possibly predicting breeding values.

Accelerating Average Information REML: An algorithm using incremental model for (co)variance component estimation in the genomic Era

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Estimation of (co)variance components (VC) is vital for genetic evaluation. The REML methods typically require the inverse coefficient matrix of the mixed model equations (MME). With genomic information, the coefficient matrix of the MME becomes denser, introducing a challenge for analysing large-scale genomic data. While the average information REML (AI-REML) is known for its fast convergence, its computational intensity poses limitations. In particular, the AI-REML requires solving the MME for each VC, which can be computationally demanding with many VC. Here we (1) present a computationally efficient algorithm, termed augmented AI-REML, which streamlines the process by solving an augmented MME only once; and (2) implement this approach under a general framework of a multi-trait GBLUP model with the general means and genetic effects. Data encompassed five traits, 12,200 phenotyped and genotyped individuals with 54,000 markers. The VC estimations were investigated based on the number of VC in the model, including 3-trait, 4-trait, and 5-trait GBLUP model, representing 12, 20, and 30 VC, respectively. We compared the augmented AI-REML with the standard AI-REML in terms of computing time per iterate. Identical VC were obtained with the same number of iterates for both AI-REML. The augmented AI-REML showed significant improvements in computational efficiency, with elapsed times of 897, 1788, and 3188 seconds for 3-trait, 4-trait, and 5-trait GBLUP models, respectively, compared to 968, 1938, and 3295 seconds for the standard AI-REML. These findings demonstrate the potential of the augmented AI-REML as an appealing solution for large-scale VC estimation in the genomic era.

A sizable putative inversion detected in the chicken genome

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Intra-chromosomal rearrangements such as inversions are common structural variations in Phasianidae birds. By analyzing 878 whole genome sequences from different species and sub-species of junglefowl and domestic chicken, we report a sizable rearrangement in the genome of chicken localized on chromosome 1 that spans over ~1.8 Mbp with coordinates at 73.6-75.4 Mbp. We explored the genome-wide allelic distribution in multiple populations of wild and domestic birds and detected a locus with a markedly different pattern of nucleotide diversity in all analyzed species. The local polymorphism content (counted as the number of SNPs observed) is ~10-fold lower in comparison to the genome-wide average. Moreover, the allele frequency spectrum within the boundaries of the locus shifts sharply in favor of certain frequency categories, denoting a local breakdown of recombination in the past. As a result, only certain classes of allele frequencies have been segregating in different populations. While the real nature of the locus is to be investigated, the variation footprint suggests that a putative inversion occurred primarily in junglefowl and passed onto the modern breeds. The region has a gene-rich sequence with a partly repeated structure that might have triggered the incidence of the rearrangement. Further analyses based on long-read sequences of wild against domestic birds are underway to reveal the real nature of the locus and possible adaptive advantage it may had on chicken domestication.

Uncovering the architecture of production-driven introgression in Cinisara cattle breed

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Local livestock breeds play a pivotal role in maintaining agricultural sustainability, biodiversity conservation, and cultural heritage. They often possess unique genetic traits tailored to local environments including rusticity, heat tolerance, and digestive efficiency. Cinisara is a dual-purpose local cattle breed of Podolian ancestry mostly farmed in western Sicily, Italy, and is an integral part of Sicilian agriculture. However, crossbreeding with cosmopolitan breeds aimed at improving meat yield has been recently implemented. To better understand the conservation status and ongoing selection of this unique breed, we investigated the current depth of Limousin introgression in Cinisara through local ancestry inference (LAI). We genotyped 30 unrelated Cinisara cattle at 65k SNPs, subsequently merging the dataset with genotype data of publicly available Italian and cosmopolitan cattle breeds. Quality checks were performed and global ancestry tests applied to determine the reference panel to be used for LAI analysis. Global ancestry confirmed the Podolian component of the breed and a pervasive introgression from Limousin. We identified multiple genomic regions showing high to complete substitution with the Limousine genome, mostly overlapping known QTLs for body weight. These preliminary results raise a warning flag on the preservation of traditional breeds, livestock biodiversity and its cultural and economic significance.

The deconvolution of bulk rat brain cell types using scRNA-seq data as reference

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The deconvolution of scRNA-seq involves estimating the cell type composition and abundance within a heterogeneous cell population based on the gene expression profiles obtained from individual cells. Huang et al., 2021 assume Seurat, SingleR, CP, RPC and SingleCellNet are the most popular and best-performing methods. Moreover, the deconvolution could also include the bulk transcriptomics data (RNA-seq) that use scRNA-seq data as a reference to perform best (Avila Cobos et al., 2020). The study aimed to deconvolute transcriptomics data based on scRNA-seq references (rat, mouse and human). As a bulk RNA-seq dataset, we used 36 brain rat samples represented by male and female animals. The brain tissues were obtained from the prefrontal cortex. The rats were supplemented with 1000 and 5000 U/Kg of vitamin D, and the unsupplemented group were control. Our pipeline focused mainly on two R packages, MuSiC/MuSiC2 (Fan et al., 2022) and DeTREM (O'Neill et al., 2023). The proportion of various cell types identified for the bulk dataset differs across scRNA-seq references. The identified cell types were mainly performed across astrocytes, GABAergic neurons, microglia, neurons, OPCs and stem cells with neuro-immune interactions.

Genetic structure and hybridization between red and sika deer in northern Italy

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Since 2004-2005, events of introgression and hybridization have been reported among red deer (*Cervus elaphus*) and the sika deer (*C. nippon*) in Northern Italy. Aim of this study was to assess the genetic structure and hybridization between these two species. The genomic DNA from 71 legally hunted animals were genotyped using the ddRAD-seq technology. A medium-density SNP genotypes panel with about 8K markers was obtained. Genetic relationships, multidimensional scaling and Admixture revealed a notable degree of admixture, indicating that hybridization was occurring between sika and red deer. These findings underscore the complex dynamics of introgression and gene flow between these two species. Hybridization can have a large effect on the genetic structure of native populations, therefore understanding admixture patterns is vital for conservation to safeguard the genetic integrity of these species.

Influence of inbreeding on fertility traits in Braunvieh and Original Braunvieh cows

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In the present study, the relationships between the inbreeding coefficient and first service conception rate (FSC), interval from first insemination to conception (FIC) and number of inseminations (NI) were analysed in Braunvieh (BV) and Original Braunvieh (OB) cows. For the study, the breeding association Braunvieh Schweiz provided fertility, milk production and pedigree data for a total of 475,330 dairy cows (BV: 453,295; OB: 22,035) with a total of 1,250,990 standard lactations (calving years 1993-2022). The mean pedigree completeness of the first 6 generations was between 90% and 100%. The mean inbreeding coefficient was 6.7%. A mixed linear model (FIC) and generalized linear models with Bernoulli (FSC) and Poisson (NI) distribution were used for the investigations. The models included the age of the cow as covariable, categories of milk yield, inbreeding coefficient and breed as fixed effects, as well as farm, year of calving, cow and residual as random effects. The FIC was log-transformed to base e for the analyses and 1 was subtracted from the number of inseminations so that there were zeros for the Poisson distribution. Inbreeding was divided into the categories 1: 0%, 2: 0-3.125%, 3: 3.125-6.25%, 4: 6.25-12.5%, 5: 12.5-18.25% and 6: >18.25%. All three traits showed that fertility decreases with increased inbreeding. Animals with an inbreeding coefficient of >18.25% have a significantly longer FIC of 65.39 days than animals in lower inbreeding categories (1: 58.64, 2: 58.87, 3: 59.41 and 4: 59.65 days). There is also a significant difference in NI between the two lowest inbreeding categories and the four upper categories (1: 2.08, 2: 2.07, 3: 2.20, 4: 2.28, 5: 2.39, 6: 2.33 inseminations). Cows with 6.25-12.5, 12.5-18.25 and >18.25% inbreeding have a significantly lower probability of FSC (0.26, 0.25 and 0.24 probability) than cows with lower inbreeding coefficients (1: 0.32, 2: 0.32, 3: 0.28 probability). The results are consistent with other studies and show that inbreeding influences the fertility of cows.

Phenotypic characterisation of Sardinian local cattle breed

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Autochthonous breeds conservation projects provide the evaluation of the usefulness of some phenotypic traits for their characterisation and valorisation. For three autochthonous Sardinian beef cattle (Sarda, Sardo Bruna, Sardo Modicana) the following traits were examined: height at the withers (HW), chest height (CH), chest circumference (CC), croup length (CL), croup width (CW), measured in cm and croup inclination (CI), lumbar back line (LL), shin thickness (TS), front stances (FA), rear stances (RA), udder volume (UV), buttock convexity (CB), temperament (T), docility (D), body condition score (BCS), nipple size (NS), nail shape (NA) measured by a score from 1 to 5. Data from 2,289 females older than 650 days (410 Sarda, 1,625 Sardo Bruna and 254 Sardo Modicana) were analysed using R statistical software using a pairwise correlation, considering the genetic types together and separately. A PCA was subsequently performed. The results showed that there were strong correlations between the parameters analysed. In particular, among the parameters with linear measurement, the highest values were highlighted, for example, between HW and CC (+0.73), between HW and CL (+0.70), between CH and CW and between CC and CL (+0.68). Positive values were also highlighted among the parameters measured with the score, while the behavioural characteristics either did not highlight particular correlations with the other parameters or showed negative values. The first two PCs explained about 55% of data variability. The variables with the greatest contribution were CB, BCS and TS for the first component and CL, HW and CC for the second. The PCA led to a partial discrimination of the three genotypes.

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Poster 27

Dissecting the genetic basis of pigmentation anomalies in flatfish through imputed genotypes

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Pigmentation anomalies present a significant challenge in flatfish aquaculture, leading to a notable decrease in the commercial value of affected fish. To address this issue, implementing marker-assisted selection or genomic selection may have great potential for enhancing the sustainability and profitability of the industry. Under this context, genomic data can be used to identify individuals with a reduced predisposition to these pigmentation anomalies. This information may enable breeders to selectively propagate fish displaying desirable phenotypes, ultimately diminishing the occurrence of pigmentation anomalies in subsequent generations. Our objective in this study was to investigate the genetic basis of pigmentation anomalies in turbot, including both pseudoalbinism (a partial or total absence of dark pigmentation on the ocular side) and ambicolouration (total darkness on the blind side), through the estimation of heritability and genome-wide association studies (GWAS). A cohort of 783 individuals (half normal and half with altered pigmentation patterns) from 10 families were genotyped using a 5K low-density SNP panel. Preliminary results revealed that the heritability of this disorder was 0.65 ± 0.09 . GWAS identified two potential candidate SNPs on chromosomes 10 and 15, respectively, which explain, however, a low proportion of the genetic variance. The availability of whole-genome data from the parental cohort, will allow imputation of low-density genotypes to whole-genome sequences and provide powerful information to deeply investigate the genetic basis of this trait. Our findings suggest that normal pigmentation can be included as a selection target in selective breeding programmes to enhance this commercially important trait in turbot aquaculture.

DHGLMF90: a tool for estimating genetic heterogeneity of residual variance using the double hierarchical generalized linear models

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In animal breeding research and applications, there is an interest in selecting uniform animals expressing a consistent response to the environment. This could be achieved by modeling heteroskedastic residuals when estimating breeding values using a hierarchical single-trait model that includes a mean and a dispersion part. The latter could be influenced by genetic and non-genetic features. Bayesian methods can estimate heteroskedastic residuals using the Metropolis-Hastings algorithm, which is slow and inefficient. Double hierarchical generalized linear models (DHGLM) represent a faster counterpart to Bayesian methods. We aimed to develop a software named DHGLMF90 to implement DHGLM with an efficient algorithm to estimate heterogeneous residual variances and breeding values for large datasets. We improved the reweighted least squares algorithm (IRWLS) to have better convergence properties than previous implementations. IRWLS iteratively calculates variance components and leverages for a bivariate model including the mean and dispersion part of the original model. DHGLMF90 uses BLUPF90+ to estimate the variance components using REML. We tested the software using simulated data and data from previous studies. We did not observe significant differences from the estimation of previous studies for most of the datasets. DHGLMF90 converged in order of magnitude faster than Bayesian methods. Further developments include analyzing datasets with genomic information and multiple-trait models.

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Poster 29

Influence of inbreeding on semen parameters in Swiss pig breeds

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The objective of this study was to explore the effect of inbreeding on semen production traits ejaculate volume, sperm concentration and number of semen portions in the Swiss pig breeds Swiss Large White (SLW), Swiss Landrace (SLR), PREMO® (PR) and Duroc (DU). For this purpose, the semen production traits from 821 boars (SLW=243; SLR=49; PR=437; DU=92) with 48,175 records (SLW=6,804; SLR=1,880; PR=34,356; DU=5,135) were used. The data were collected in the years 2009-2016. The mean pedigree completeness of the first 6 generations was between 90% and 100%. Mixed linear models were used for the investigations. The model included the fixed effects birthyear of boar, age of boar x month of collection, year of collection, collection interval, inbreeding coefficient, breed, and boar handler as well as random animal and residual effects. The sperm concentration was used in ln-transformed and the number of semen portions in square root transformed for the analyses. Inbreeding was divided into three categories 1: 0-3.125%, 2: 3.125-6.25, 3: 6.25-12.5%. Inbreeding depression was evident in the traits ejaculate volume and number of semen portions, but not in sperm concentration. Animals in inbreeding category 1 had a significantly higher volume of 274 ml than those in categories 2 (267 ml) and 3 (265 ml). In terms of concentration, animals in inbreeding category 3 had a slightly lower concentration of 263 Mio./ml than those in inbreeding categories 1 (270 Mio./ml) and 2 (271 Mio./ml), but the difference was not significant. Significantly fewer number of semen portions were obtained from animals in inbreeding category 3 than from those in the other two inbreeding categories (1: 38.7, 2: 37.9, 3: 36.4). The results are consistent with other studies, showing that the inbreeding influences the boar fertility. The effects of inbreeding reported in this study were small and can be compensated by additive genetic breeding progress.

Multicriteria performance of five grass-based cattle farms along a gradient of stocking rate

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The debate on the sustainability of livestock farming is often polarized around greenhouse gas (GHG) emissions and land use, and the potential benefits of grassland-based ecosystem services in farm assessments are generally overlooked. Accounting for multiple ecosystem services (ES) may be further complicated by confounding effects of animal stocking rate and trade-offs between ES. We used life cycle assessment to assess environmental impacts of four “green options” for cattle production and a conventional dairy farm that are distributed along a broad gradient of extensification in the Atlantic area of Western Europe. We also applied an ES-multifunctionality assessment method to these five farms in which multifunctionality was defined and valued according to different stakeholder perspectives. We showed that relying on C sequestration in grasslands to fully compensate for ruminant GHG emissions would lead to farming at a very low stocking rate. The climate-neutral farm had 0.53 livestock units/ha of on-farm fodder area and produced 11 kg of human-edible protein (HEP)/total ha.yr. The sustainable intensification farm produced 124.5 kg HEP/total ha.yr and was also climate-neutral. Following a “land sparing” strategy, high-yielding cows grazed temporary grasslands and annual fodder crops, while carbon was sequestered in soil and woody biomass on the half of the farm area that was “returned to nature”. The types of biodiversity on these five farms differed as well as their cultural value. We discuss how the inclusion of stakeholder values in multi-criteria farm assessment provides valuable information for the systemic redesign of cattle production systems.

Session 73

Theatre 2

Agroecological transition may reduce the fat content of Fourme de Montbrison cheese

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The Fourme de Monbrison (FM) is a blue type PDO cheese from the French Massif Central area made with unskimmed milk. Since 2011, the dairy farmers are moving towards more agro-ecological practices, promoting in particular a transition from form Holstein to Montbéliarde (Mo) breed and from maize silage to herbage as forage base, maximizing grazing. In parallel, since 2018, the share of cheeses having a low fat in dry matter (F/DM) content (< 52% which is the minimum threshold in the specifications of FM cheese) increased and reached 10% and the milk fat content decreased by -0.25 g/kg milk per year, especially at the beginning of the grazing period. An experiment was set up in 20 FM farms to monitor the evolution of milk fat and protein contents and fat to protein ratio (FPR) during the grazing season, in relation to farming practices and herbage characteristics. Milk fat was negatively related to pasture energy content and organic matter digestibility ($R < -0.37$) and positively to its fiber contents ($R > 0.40$). Milk protein was positively related to pasture protein content ($R > 0.35$) and negatively to the time elapsed since the beginning of the grazing season ($R = -0.40$). This suggest that grazing at an early vegetative stage can depress milk fat content and FPR and therefore cheese F/DM content. The proportion of Ho in the herd was positively related to FPR ($R = 0.33$) and negatively to milk protein ($R = -0.57$). In order to tackle these issues, some farmers are willing to test corrective practices in their own farm in the frame of the EU H2020 INTAQT project's “living-labs” activities.

Meat production from organic suckler cattle fed on grass

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Suckler cattle struggle to find their place in organic farming: they are poorly represented in the beef sector because of the lack of market for young grazer and of price differentiation with the conventional market and technical difficulties related to the lack of organic fattening benchmarks. This study was performed at INRAE's facility (Herbipole) in Laqueuille (Puy de Dôme, France). The aim was to position medium-mountain suckler cattle in the organic meat supply by proposing new production itineraries for young males, as alternatives to the current lean or beef schemes. In this experiment, 29 young males Salers cross Angus were produced in mountainous areas (altitude 1000-1500m), 12 in 2021 and 17 in 2022, and slaughtered at less than 12 months of age, 14 of them were castrated (C). Mother-calf pairs were fed without concentrate until weaning. The males were fattened for around 3 months on a ration based on natural grassland haylage, supplemented with grain meslin at $\pm 30\%$ of total ingested dry matter of ration. Calf growth during suckling period was 1200 g/d, thanks to the good milk production of the dams and quality of the pasture. During fattening, growth was ± 1380 g/d, with a significant difference of 300 g in favor of uncastrated calves (UC). Carcasses were heavier for UC than for C: 280 kg vs. 255 kg. Crossbreeding with the Angus breed and castration produced carcasses with good fat cover (BCS 3-3.5), carcass fat and marbling were satisfactory for young males under a year old. This study showed that fattening young males with grass-based diets and few concentrates in accordance with the organic specifications is possible by combining genetic type, good pasture and forage quality. This production also meets societal demand and sustainability.

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Theatre 4

Predation on cattle: between the wolf's responsibility and the absence of damage reports

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Over the past two decades, wolf predation on cattle farming systems has surged. A analysis of damage reports was carried out in Savoie and Haute-Savoie (147 reports, 2015-2020) to mitigate impacts. An online survey was also conducted on 1591 cattle farmers to get a better understanding of the situation on the ground (673 responses, 35.5% of cattle farmers in the 2 Savoies). Findings highlights the difficulties in reaching conclusions from the expertise: 37% of the damage was determined to be unrelated to wolf predation, while 63% indicated the possibility of being attributed to potential attacks. However, in the majority of both cases, a cause of death of undetermined origin was identified: 56% for non-wolf damages and 58% for potential attacks. Then, only 10% of cattle farmers report damage, while 43.5% experiencing unreported losses. Among them, 46% reported missing animals. Cattle rockfall, mortality, lameness, fractures, and atypical injuries affected 29%, 26%, 17%, and 10% of the farmers, respectively. Plus, 68% of farmers didn't reported their damage because they didn't know whether it was wolf-related, and 28% didn't know how to report it. Only 20% knew the entire reporting process (40% no knowledge, 40% moderately familiar). The lack of proof and the challenges of determining the wolf's responsibility, along with procedural unfamiliarity, leads to an underestimation of the damage. Therefore, procedures and information require review and adaptation. This study prompts adaptation strategy identification to reduce risk, impacting grass-fed cattle farming areas.

Ways to achieve protein autonomy in cattle feeding

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Nitrogen (N) is a vital element for plants, animals, and humans. However, reactive N compounds (Nr) such as NH₃, NO₃⁻, and N₂O can harm animals, humans, and the environment. Approximately one third of anthropogenic Nr losses may be associated with the production of food of animal origin. The main N inputs to farms are from protein-containing feeds, N fertilisers, symbiotically derived N, and atmospheric N deposition. In turn, N is exported from farms via the products feed, milk, and meat, but Nr losses also occur. Such losses are strongly related to the N input–output balance and thus to imported N inputs via feeds and fertilisers. Therefore, one way of reducing N losses is to increase the N autonomy of farms, thereby decreasing the dependence on imported N through feeds. In cattle farming, N autonomy can be improved by increasing the proportion of protein-rich roughage in the ration. Using legumes in grass stands may increase the N yield of herbage and limit the use of N fertiliser. Lowering N losses during the conservation of forages also reduces the dependence on N inputs. Home-grown grain legumes could also improve N autonomy, but this raises issues of protein yield compared to legume-rich leys and specific climatic conditions. Feeding also offers a multitude of opportunities to improve N autonomy, for example, through more precise feeding recommendations, feed additives, treatments of protein-rich feedstuffs, and protein-reduced feeding. The use of cows adapted to herbage-based, low-input feeding systems and breeding for N-use efficiency may be other practices for increasing N autonomy. All these measures have different impacts on different environmental metrics, productivity, and product quality, which need to be weighed up.

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Theatre 6

Effect of concentrate crude protein and amino acid supplementation on milk production of mid-lactation, grazing dairy cows

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The objective of this experiment was to investigate the effect of concentrate crude protein (CP) and amino acid supplementation on milk production in mid-lactation, grazing dairy cows. One-hundred mid-lactation Holstein-Friesian cows were blocked based on pre-experimental milk production and parity and then randomly assigned to 1 of 4 dietary treatments (n=25). The treatments were pasture supplemented with concentrate comprising of either; 1) 17% CP (H); 2) 13% CP (M); 3) 9.5% CP (L); or 4) 9.5% CP with rumen-protected amino acids (L-AA; 8.0 g/day absorbable Met and 7.2 g/day absorbable Lys). Concentrate CP concentrations are reported on a dry matter (DM) basis. The concentrates were fed at 1.78 kg of DM/cow/day in the milking parlour over an 8-wk experimental period. All data were analysed using PROC MIXED in SAS. Cows fed M had similar milk yield when compared with H and L but were higher than L-AA (20.4, 20.2, 20.0 and 19.7 kg/day, respectively; P<0.05). Fat concentration was similar for cows fed M, H and L-AA but all treatments were higher than L (4.95, 4.82, 4.95 and 4.65%, respectively; P<0.01). Cows fed L-AA had higher milk protein concentration when compared with L but were similar to M and H (3.91, 3.81, 3.88 and 3.84%, respectively; P<0.01). Cows fed M had similar milk solids yield when compared with H and L-AA but were higher than L (1.79, 1.73, 1.74 and 1.68 kg/day, respectively; P<0.01). These results encourage producers to reduce concentrate CP to 13% of DM when supplementing mid-lactation grazing dairy cows; however, if reducing to 9.5%, rumen-protected amino acids should be considered.

Green-cut sorghum fed indoor to dairy cows: a strategy to mitigate herbage shortages?

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This study compared the effects of feeding lactating dairy cows with a green-chopped multi-cut Sorghum x Sudangrass hybrid, with or without addition of sulphur, or freshly harvested herbage from a grass-clover mixture. Sorghum was sown beginning of June 2023 in Posieux (600 m a.s.l.). Twenty-four Holstein cows (194±31 d in milk, 26.0±4.0 kg/d milk yield, 638±53 kg body weight) were blocked by lactation number, days in milk, and milk yield, and randomly allocated to 1 of 3 diets (sorghum, sorghum+sulfur, herbage) of 8 cows each. From end of August, during 4 weeks (2 weeks adaptation, 2 weeks sampling), the first regrowth of sorghum (mean height of 1.2 m) and a 4th regrowth of herbage were harvested daily and fed ad libitum from 0700 to 1700h in automatic roughage intake control bins. Overnight, individual access to maximum 10 kg DM hay was provided. In addition, 1.9 kg DM/day on average of a concentrate feed (6.7 MJ NEL, 12.3% CP) including a mineralized and vitaminized premix (with or without calcium sulphate dihydrate) and bait feed in a GreenFeed were fed. Feed intake averaged 21.3±0.5 kg DM/d (mean±standard error) and did not differ among groups. Fresh forages supplied 48±2.2% and 65.0±2.4% of total DM intake in sorghum- and herbage-fed cows, respectively. Milk yield averaged 22.7±0.7 kg/d and did not differ among groups. Milk fat and protein concentrations did not differ among groups either, whereas milk urea was higher (+15 mg/dl) in herbage- than in sorghum-fed cows. These preliminary results show potential for green-cut sorghum to alleviate herbage shortages in late summer. Additional data on methane emissions and mineral status are currently being evaluated.

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Theatre 8

Effect of dietary forage proportion and crossbreeding on dairy cows' feeding efficiency and methane emissions

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The aim was to assess the effect of forage proportion (FP) and crossbreeding on feeding efficiency and methane (CH₄) emissions. Calorimeter chamber data (n=908) obtained from 32 experiments with Holstein-Friesian (HF; n=796), Norwegian Red (NR; n=50), Jersey x HF (J×HF; n=46) and NR×HF (n=16) were split into FP groups (% DM basis): Low (LFP, 10-30%), Medium (MFP, 30-59%), High (HFP, 60-87%) and Forage-Only (FOR, 100%). Data were analysed using a linear mixed model with FP and breed as fixed factors, and animal and study as random factors. Multivariate redundancy and regression analysis further drew correlations between drivers (diet, breed) and dependent variables (efficiency, CH₄). Milk yield (kg/d; P<0.001), energy-corrected milk yield/dry matter intake (ECMY/DMI; kg/kg; P<0.001) and milk energy/DMI (MJ/DMI; P<0.001) were higher in LFP (25.0, 1.43, 4.43) and MFP (22.6, 1.32, 4.10) than in HFP (17.5, 1.20, 3.72) and FOR (11.9, 0.89, 0.95, 2.93). CH₄/DMI (g/kg) was higher (P<0.001) in HFP (24.3) than in MFP (22.4). CH₄/ECMY (g/kg) was higher (P<0.001) in HFP (21.6) and FOR (25.8) than in MFP (17.9). When compared with higher forage diets (60-100%), low-medium forage diets (10-59% DM) improved productivity and efficiency, while medium forage diets (30-59%) also reduced CH₄ emissions per kg DMI and milk yield.

Emission measurements in a naturally ventilated dairy housing: How does the crude protein level of the diet affect ammonia and nitrous oxide emissions?

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Swiss farms are encouraged to maximise milk production by using homegrown forage. However, a high proportion of grass-based feed components such as grass silage, hay, or fresh grass in the diet can lead to a protein surplus. The aim of this study was to measure emissions of ammonia (NH₃) and nitrous oxide (N₂O) from two typical Swiss dairy cattle diets with varying crude protein (CP) levels. NH₃ and N₂O emissions were quantified in two separate housing compartments each for 20 lactating cows over six days. Cows in one compartment received a diet with 116 g CP kg⁻¹ dry matter (DM), while cows in the other compartment were fed a diet with 166 g CP kg⁻¹ DM. The diets were then switched for a second measurement period. A tracer-ratio method was used to measure emissions under natural ventilation. Linear mixed effects models identified temperature ($p < 0.001$), wind speed in the housing ($p < 0.001$), and the diet ($p < 0.001$) as factors influencing NH₃ and N₂O emissions related to livestock unit (LU), dry matter intake or energy corrected milk. Reduction of NH₃ and N₂O emissions per LU was on average 46% and almost 20% for the low-CP diet compared to the high-CP diet. Strong correlations were observed between dietary CP and urinary nitrogen excretion ($R = 0.84$), dietary CP and milk urea content ($R = 0.82$) as well as urinary nitrogen excretion and milk urea content ($R = 0.71$). Our full-scale study showed that a significant reduction in NH₃ and N₂O emissions can be achieved by lowering the CP level of the diet.

Session 73

Theatre 10

Willow (*Salix* spp) tannin-rich tree fodder: The potential to reduce methane and improve productivity of grazing beef cattle

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Willow contains condensed tannins (CT) and could be introduced in silvopastoral systems addressing environmental problems in the livestock sector. This study aimed to explore the effects of willow CT on animal performance, emissions, and faecal metabolome. Growing beef steers ($n = 20$) were allocated in a 2 (treatment) x 2 (period) Latin square design study. Treatments were Perennial Ryegrass (PRG) vs Willow fodder mix with a grass understory (WFG) containing 45 gCT/Kg DM. A beef monitor unit recorded individual live weight (LWG). Gaseous exchanges were measured using 2 GreenFeed units and data was used to calculate heat production (HP), ME (MJ/d) and GE (GEI) intake. Metabolic shifts associated with treatment-induced differences explored with rapid evaporative ionisation mass spectrometry (REIMS) on faecal samples. The LWG (kg/d) was higher ($P < 0.01$) for PRG during P2 (1.29 vs 0.69) but HP (MJ/d) and DMI intake did not differ between treatments. CH₄ (g/d) production was reduced ($P < 0.001$) by 27% and CH₄-E/GEI by 22%, ($P < 0.001$) for the WFG vs PRG treatment, (173.5 vs 236.7) and (6.0 vs 7.7), respectively. No significant differences were observed for H₂ production (g/d). Two-dimensional PLS-DA scores plot shows clear separation between the faecal metabolome of PRG and WFG.

Virtual Fencing for alpine grazing and resource conservation: opportunities and challenges

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From an environmental perspective, alpine pastures provide multiple functions and a large number of ecosystem services, such as biodiversity conservation, prevention of soil erosion, climate regulation, and cultural services. They are also sometimes the only source of forage for summer grazing livestock. However, they are often located in inaccessible areas, leading to their gradual abandonment, with negative consequences for these resources. In this context, the use of Virtual Fencing (VF) may become a key tool for grassland conservation. VF technology is based on GPS collars capable of managing animals by setting virtual boundaries of grazing areas through the delivery of a coupled aversive stimulus: an audio cue followed by an electrical pulse. The feasibility of using VF in mountain areas was tested throughout the grazing season of 2023 in the Eastern Italian Alps (Mount Zoncolan, IT) on 35 Bruna lactating dairy cows. The animals were managed in 3 different pastures with an average altitude of 1600 m a.s.l.: two malgas, (Pozzo and Tamai) and ski slopes (using a mobile milking parlour). During the 70 days of management, the animals responded positively to the VF, with a slight increase in the emission of stimuli when managed on the ski slopes. However, on some days collars did not report any recordings due to the poor GSM coverage and quality (2G) in areas where the infrastructures level was low. Nevertheless, the study showed VF technology as a promising solution for sustainable livestock management in alpine pastures, balancing the needs of agriculture with those of environmental conservation.

Session 73

Poster 12

Effect of silage species and concentrate crude protein level on early-lactation milk production in grazing dairy cows

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The objective of this experiment was to determine the effect of silage species and concentrate crude protein (CP) level on milk production in grazing dairy cows. Eighty early-lactation, spring-calving cows were blocked based on pre-experimental milk production, parity and breed. Cows were randomly assigned to one of four dietary treatments in a 2×2 factorial design including two silage species (perennial ryegrass (PRG) silage (GS) or PRG-red clover silage (GRCS)) and two concentrate CP levels (17% (HP) or 9% (LP) of dry matter). All groups grazed separately by day and received 7 kg of dry matter/cow/day of silage at night along with 3.56 kg of dry matter/cow/day of concentrate. The study consisted of a 2-wk covariate period and five weeks of data collection. Data were analysed using a PROC-MIXED model in SAS. Compared to HP concentrate, cows fed LP had lower milk yield (24.5 vs 23.0 kg/cow/day, respectively; $P < 0.001$) and milk solids yield (1.99 vs 1.89 kg/cow/day, respectively; $P < 0.01$). Silage species had a significant effect on milk fat concentration, with cows fed GS higher than GRCS (5.07 vs 4.81%; $P < 0.01$); however, there were no differences in milk yield (23.7 vs 23.9 kg/cow/day, respectively; $P > 0.05$) or milk protein concentration (3.33 vs 3.35%, respectively; $P > 0.05$). No interaction between silage species and concentrate CP level was observed ($P > 0.05$). As PRG-red clover silage reduced milk fat production at this feeding level, future work should investigate feeding various levels of PRG-red clover silage to grazing dairy cows.

Protein self-sufficiency and nitrogen use efficiency in beef-on-dairy crossbred bulls

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Optimizing the use of feed protein by increasing farm self-sufficiency and animal use efficiency have economic and environmental benefits. Two iso-protein and iso-energy diets based on corn and grass silages, supplemented with either peas and barley or faba beans and triticale were fed ad libitum to 75 crossbred bulls (257 kg BW) from a ♀ Brown Swiss and a ♂ of Angus (An), Limousin (Li) or Simmental (Si) up to a BW of 520 kg. Post-mortem chemical analysis of two bull's empty bodies and carcasses from each crossbreed at the start of the trial and use of predictive equations from 11th rib dissection and carcass CH-TAX grading for the 69 remaining bulls allowed estimation of initial and final body and carcass chemical composition. The individual amount of protein fixed during fattening was further estimated by difference, whereas the ratio of body protein fixed to protein ingested was defined as the nitrogen use efficiency (NUE). Relationships between NUE, average daily gain (ADG), feed conversion ratio (FC: dry matter intake/ADG) and carcass composition were assessed by Pearson correlation. They were particularly suitable for the Si crossbred bulls which had the highest NUE (25.0%), followed by Li (21.4%) and lastly An (19.0%, $P < 0.001$). The NUE was positively correlated ($P < 0.001$) to the ADG (Si 1.65 > An 1.50 > Li 1.40 kg/d), to the proportion of protein in the carcass (Si 18.7 and Li 18.6 > An 18.1%) and to feed efficiency (FC: Si 4.93 < Li 5.56 and An 5.63 kg/kg). A mild negative impact on the commercial value of the carcass was observed in Si, with a grade of fat cover of 2.5, lower than the optimum CH-TAX score of 3. An overall assessment including other aspects such as meat quality or greenhouse gas emissions will enable conclusions to be drawn about the sustainability of this protein self-sufficient fattening system.

Session 73

Poster 14

Milk production of Holstein cows grazing fescue at different defoliation intensities

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The impact of two defoliation intensities on a *Lolium arundinaceum* pasture on Holstein dairy cows' milk production (MP) and composition (fat, protein, and lactose concentration; MC) was studied in Uruguay. The treatments were two residual sward height: Lax (TL) and Traditional (TT) with 12-15 cm and 5-7 cm, respectively. Thirty-two autumn calved multiparous cows were grouped in sets of four, allocated to 1.6-ha paddocks in a randomized complete block design with 4 spatial replicates. The trial began on April 19th, and the same cows were used throughout the study, managed in a rotational grazing with 6h of pasture access). During spring (August 1st to November 19th) cows consumed 11.7±1.14 kg DM/d of a partial mixed ration with a 64:36 concentrate:sorghum silage ratio after evening milking, in a compost barn. MP was registered automatically twice daily. Every fifteen days an individual milk sample was collected in each milking to determine MC. Data were analysed using GLIMMIX of SAS with a mixed model that included treatment, month, and their interactions as fixed effects. Pre- and post-grazing sward heights were 21.5±3.10 cm and 14.4±1.74 cm for TL, and 24.0±5.29 cm and 8.0±1.63 cm for TT; respectively. The MP was greater on TL than TT (29.7±0.96 vs 27.0±0.90 kg/cow/day; $P = 0.019$) while MC was not affected by the treatments (3.8±0.08% fat, 3.4±0.42% protein, and 4.8±0.27% lactose; $P = 0.928$). The MP decreased 8.6±0.15 kg/cow/day during the study ($P < 0.0001$) with larger differences between treatments in November (26.1±0.98 vs 21.9±0.91, TL and TT, respectively), suggesting effects of grazing intensity on lactation persistency. Lax management produced more milk than traditional with no changes in MC.

Meat quality of extensively reared beef cattle in Greece

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The aim was to assess meat quality from beef reared extensively in the natural grasslands of Axios River delta in Greece. The study farm comprised 200 crossbred beef cattle with an average age and body weight at the slaughter of 23±1.0 months and 586±11.54kg. A total of 22 meat samples (9th rib) and 15 samples of plants during the grazing period were collected and subjected to chemical analyses and quality assessment. Results of texture parameters including Hardness1, Hardness2, springiness, cohesiveness, chewiness were 1896.9±1484.75g, 846.6±337.32g, 0.7±0.11%, 0.5±0.09%, and 395.2±161.3g, respectively. Colour values for Lightness—L*, Redness—a*, and Yellowness—b* were 36.7±2.53, 17.7±2.02, and 8.3±1.41, respectively. Average fat and protein content was 1.1±0.79% and 22.9±0.83%, respectively. In plants, concentrations of oleic and linoleic acids were 10.5±5.27% and 21.5±4.02%, respectively. Likewise, concentrations of prior mentioned acids in beef were 38.9±5.85% and 3.1±4.76%, respectively. A notable concentration of linolenic acid (20.0±11.95%) was reported in plants, while in beef, low levels of palmitic (22.8±4.90%) and myristic (2.1±0.32%) acids were observed. Plants' polyphenol content was 63.4±58.95mg/kg, mirroring the richness observed in beef (17.7±4.05mg/kg). Findings underscore the benefits of natural grazing in meat quality. This work is funded by "Axios Meat" project "Measure 16 'Cooperation" (Project code M16SYN2-00025).

Toasted Faba Bean and wheat mixture as an alternative protein source in a low protein diet for dairy cattle

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The high rumen protein degradability of field beans (FB) render them inferior to soybean meal (SBM) for high producing dairy cattle. However, toasting FB offers an economically attractive option as a meta-analysis revealed a reduction in ruminal degradation of both protein (13g/100g) and starch. Further, intercropped field bean-wheat (FBW) post-toasting emerge as a promising local protein and energy source for organic dairy farmers, certainly when minimizing protein use. The aim of this trial was to investigate the inclusion of FBW in a low protein diet (14.5% crude protein) of high producing dairy cattle on performance and N efficiency. A trial was set up with 2×17 high producing dairy cows (204±9 DIM, producing 34.0±0.8kg of milk). A basal diet consisting of maize silage, prewilted grass silage and pressed beet pulp (52/37/11 on DM) was fed ad libitum. The control diet was supplemented with 6.1% SBM and 0.23% feed urea, whereas the experimental diet with 6.8% FBW, 2.3% SBM and 0.41% feed urea. Further both groups received equal amounts of rumen protected SBM and concentrates to meet 105% of the energy and protein requirements of each cow and to attain a rumen degraded protein balance of 40 g/d. Dry matter intake (DMI), milk yield (MY) and milk composition were compared using ANOVA (p<0.05). Total DMI was not different between CTRL (24.8±0.4kg) and FBW (24.9±0.4 kg) and also MY (32.2±0.7 vs. 31.9±0.7kg), milk fat (4.65±0.09 vs. 4.70±0.09%) and milk protein (3.87±0.05 vs. 3.87±0.05%) were not affected. Nevertheless, N efficiency (N-output in milk/N-input by feed) was better (p<0.05) for FBW (32.8±0.4%) compared to CTRL (31.8±0.4%). In this trial FBW successfully replaced part of the SBM in a low protein diet.

Session 74

Theatre 1

Animal Welfare and Animal Production Systems, the Case of Horses

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Increasingly, and across many forums, the issue of Horse Welfare is being developed into new models, and as such, is receiving greater and greater attention. One aspect seldom explored, however, is the role of Animal Production Systems in the ‘production’ of Horse Welfare. This presentation will focus on this, examining the different meanings and definitions of Horse Welfare from the perspective of Horse Production Systems. It will examine the potential impact on horses of different uses and different aspects of the system in an attempt to bring a more detailed and nuanced understanding of Horse Welfare as the Horse Production System moves forward in the 21st Century.

Session 74

Theatre 2

Mind the Gap: Leveraging Human Behaviour Science to improve Animal Health and Welfare

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In order to improve animal health and welfare, it is nearly always necessary to work alongside the human animal, encouraging other humans to change their behaviour. Understanding the science behind human behaviour change is an essential component in facilitating better animal well-being, yet is frequently overlooked in animal welfare studies. This talk delves into the critical nexus between human behaviour and animal health, elucidating why a deeper comprehension of people is pivotal. Drawing upon interdisciplinary insights from psychology, sociology, and behavioral economics, we unravel the underlying mechanisms shaping human actions and decisions, and their profound impact on animal welfare outcomes. At the heart of this thesis is the recognition that it is human behaviour which determines the success or failure of animal health interventions. Whether encouraging compliance with treatment regimens, fostering responsible pet ownership, or promoting sustainable agricultural practices, our ability to effect positive change hinges upon our understanding of human behaviour dynamics. In this talk, we explore the principles of behavioural science, enabling us to design more effective interventions and communications in order to cultivate lasting behaviour change among stakeholders. Through case studies and empirical evidence, this presentation explores the imperative for a paradigm shift in our approach to animal health and welfare. By integrating insights from human behaviour science into our toolkit, we not only enhance the efficacy of our interventions but also foster a deeper appreciation for the interconnectedness of human and animal welfare.

The end of life of equidae in France

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In France, socio-economic studies carried out in the equine sector have established that over the period 2010-2021 the population of equidae aged over 20 has increased by more than 30%. In 2010, equidae aged over 16 represented just 18% of the French equine population, whereas by 2021 they accounted for 28%. Over the same period, the number of slaughtered horses has fallen by 77%. These statistics highlight two types of change in the relationship between humans and equidae: on the one hand, longer animal careers that include retirement, and on the other hand, new approaches about equidae end of life. What are the determining factors of these trends? What would make a good end of life for equids in France today? This talk relies on the results of two research projects carried out between 2017 and 2021. The first one questions living and working conditions between humans and horses through 108 semi-structured interviews, whilst the second one focuses on animals leaving a working life thanks to 20 qualitative surveys. Results are analysed using the theoretical framework of animal work sociology. The talk will begin by reviewing the history of horse slaughter in France from the 19th century to the present day. The aim is to show how the promotion or condemnation of equine slaughter is based on moral considerations towards animals. The second part will present the results of the two studies. Professionals from the equine sector highlight three different ways in which the lives of equidae can be ended: slaughter, euthanasia and natural death. The notions of a good death compared to a beautiful death will be analysed. The particularity of working with domestic animals lies in the fact that humans enter into a tacit contract of gift – against gift with their animals. In other words, the gift that animals give to humans by responding to the tasks they ask of them, who in return undertake to provide them with protection and care. This moral commitment of humans towards their animals is exacerbated at the end of their lives. The final part will therefore look at the moral tensions that professionals feel towards their animals and the management of their end of life: sending them to the slaughterhouse, when to euthanize, managing the dead body (between disgust with the rendering service and legal obligations).

Session 74

Theatre 4

Enhancing Health and Welfare of Non-for-human-consumption (non-DPA) Horses: An Online Investigation

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In Italy, equids are mandated to be registered on the Equine Data Bank (BDE), where owners categorize them as either "non-dpa" (not for human consumption) or "dpa" (intended for human consumption). Regulations prohibit the slaughter of retired athletic horses, posing challenges in their post-career phase. This study aims to improve the management of non-dpa horses by assessing the current practices and awareness levels. An online questionnaire was developed in a prior project, covering legal aspects, breeding, welfare during sports and transport, and the crucial "end of life" phase. The initial survey revealed a lack of representation from the horse racing sector, prompting the need for further investigation, particularly into the challenges faced during the "end of career" phase. Therefore, a modified questionnaire was designed to include a section on biosecurity, acknowledging the frequent movement of non-dpa horses for various purposes. The objective is to encourage the adoption of biosecurity plans and enhance awareness among owners regarding disease prevention. Although online survey results are not available yet, previous project feedback suggests re-emergence of logistical issues managing non-DPA horses at the end of career. While many know common biosecurity rules like vaccinations, few are aware of less common measures like PPE use in horse facilities. Collected data will provide official control indicators. The results will be instrumental in developing a checklist for competent authorities, ensuring compliance during inspections. Ultimately, this research aims to enhance the knowledge and competence of horse owners, managers, and operators, fostering better practices for the welfare of non-dpa horses.

Swedish dairy farmers' and trotting-horse trainers' perceptions of animal welfare inspections

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To ensure proper compliance and enforcement with animal welfare (AW) regulations some kind of control system is necessary. In Sweden, the County Administrative Boards (CABs) perform the official AW inspections. However, there are also private AW standards linked to separate audit systems. Hence, the control arena is complex with different AW inspections taking place at farm level. This study investigated dairy farmers' and trotting-horse trainers' views on different AW inspections. A digital questionnaire was distributed in which the dairy farmers were asked about their perception of inspections from CAB, Arlagården® and KRAV, and the trotting-horse trainers were asked about inspections from CAB and the Swedish Trotting Association (STA). A total of 216 farmers and 396 trainers responded. Data were first compiled using descriptive statistics and then correlations were applied when relevant. In general, the farmers more often reported being worried before an AW inspection, e.g. 60% of the farmers stated to be worried before a CAB inspection compared with 10% of the trainers. The farmers also had a more negative view of the CAB inspections compared with the trainers, with 37% of the farmers having a negative experience compared with 10% of the trainers. However, the trainers reported significantly more positive experiences from the STA inspections than for the CAB inspections. Knowledge about the trotting horse business and dairy farming respectively were the most important traits for inspectors, according to trainers and farmers. It was also considered important that inspectors could make uniform, but flexible, assessments. Other factors associated with the experience were e.g. the outcome of an inspection, age and gender of the inspector, and if the inspection was pre-announced or not, especially for the farmers. In general, the trotting-horse trainers had a more positive view on being inspected compared to the dairy farmers, who had a more diversified view.

Session 74

Theatre 6

Horse welfare in semi-extensive system: the case of the Italian Catria horse breed reared for meat production

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Catria horse is an Italian breed reared for meat production in semi-extensive systems, on pastures during the warm season and on stable during the cold season. A welfare assessment protocol for horses suitable to be applied on pasture and on stable is still not available. The aims of the study were (i) to develop and apply a welfare protocol that considers non-animal-based measures and animal-based measures (ABMs); and (ii) to evaluate whether the welfare items were influenced by the breeding system (pasture vs stable). A total of 65 welfare items, classified into 6 thematic areas (training, feeding, facilities, ABMs, biosecurity, health management) were selected by a focus group. The protocol was applied by a trained veterinarian on 429 horses dislocated across 26 pastures and 7 stables. Data analysis was performed by Python packages ('SciPy' v1.2.1) according to Fusi et al. [1]. Statistical significance was $p < 0.05$. The differences between pasture and stable were related to the 'training' ($p = 0.02$) and the 'feeding' ($p < 0.01$) areas. The pasture received a higher frequency (%) of inadequate answers compared to stable ($p < 0.01$) for the welfare items 'inspection of the animals' (no daily inspection was performed) and 'feeding management' (no rations were planned according to the physiological status of the animals). However, horses housed on stable were fed with inadequate quantity of cereals-based concentrate feeds, by overloading the safe level of starch content per meal. Weakness points for both pasture and stable were represented by some welfare items related to the health management ('health management of the foal', 'hoof care', 'dental care' and 'parasite management') and the biosecurity area ('measures of pests fighting' and 'measures for the entrance of visitors'). The study filled the existing gap on horse welfare protocol and allowed to identify the main weakness points to address preventive interventions. [1] Fusi, F. 2021.. Anim. Prod. Sci. 61, 55–63.

Does the dominance hierarchy in a large group of horses influence locomotor activity and area utilization?

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Group husbandry systems account better for the behavioural needs of horses (e.g. social contacts, movement possibilities). However, it is necessary to understand the behaviour of horses better with regard to optimally designing such systems, specifically with regard to promoting movement and diminishing negative effects for lower ranked horses. This study adds to our understanding of the interlink between rank and locomotor activity. A group of 40 horses kept together in one large group on pasture was equipped with GPS sensors from August to October 2022 to monitor the daily locomotor activity as well as the area utilization. Approximations about the dominance hierarchy in the group were made by pair-feeding tests and calculation of a dominance index based on the feeding time of the horses in this test setting. Statistical analysis was carried out with generalized linear mixed modelling, taking into account the fixed effects rank, age and test day. The horses travelled on average 5.4 ± 1.2 km/day, with no significant effect ($P > 0.05$) of social rank. However, older horses had a lower locomotor activity (age was a significant effect with $P < 0.05$). For the determination of area utilization, the available space was divided into a total of 8889 quadrants with 3×3 m each. Area utilization was then determined as how many of these quadrants each horse visited per hour and averaged 37.9 ± 6.4 , with no effect of social rank ($P > 0.05$) but a significant effect of age ($P < 0.05$) observed. The study suggests that in this specific setting, social rank did not significantly affect locomotor activity or area use.

Session 74

Theatre 8

Equids welfare in Italian traditional equestrian events: comparison between 2022 and 2023

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Traditional equestrian events, such as tournaments and races, where equids (donkeys and horses) are used outside authorised hippodromes, are still widespread in Italy and represent important religious and historical events. In compliance with Ordinance 21st July 2011 of the Italian Ministry of Health, safety of riders, equids and spectators must be ensured during these events. Since 2017, official veterinarians must complete a survey to identify hazards for the equids and report any adverse effects occurring during an event. If any hazards are identified, they can order changes or even stop the event. These data are sent to the Italian Reference Centre for Animal Welfare for surveillance and reporting. The number of equestrian events and equines involved increased in 2023 compared to 2022 (136 vs. 109 events; 2651 vs. 1754 equids). The events were more frequent during spring and summer in both years and during the carnival period in 2023 unlike in 2022 probably due to covid-19 pandemic restrictions still in place in early 2022. No differences were seen regards to veterinary and staff number, presence of fences, guards and animal shelters, which were present at almost all events. More than half of the events consisted in races which took place on city streets or squares, with the surface covered by sand and/or dirt (62.5% in 2023 and 61.5% in 2022). In 2022 one event was cancelled since the surface consisted of only cobblestone. In both years, around 85% of equids involved were horses; 44% were female and about 42% were 4-8 years old. Since animals under 4 years of age are not allowed to race, 11 equids in 2022 and 3 in 2023 were excluded from the events. Injured animals were 0.46% (with 1 dead horse) in 2022 and 0.26% in 2023 (no deaths). Considering that less animals were injured in 2023 despite more animals being involved, the survey seems to be a good tool for risk assessment and mitigation of hazards that may adversely affect animal welfare and safety.

Quantifying the effect of loading density and gender on selected blood components and carcass bruises of slaughter horses

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The objective of this study was to determine the effects of loading density (high and low) and gender (mares, geldings and stallions) on blood welfare indicators and carcass bruises of slaughter horses. Data was recorded for eleven shipments referring to 89 slaughter horses that originated from the same collecting point. Interactive effect of loading density and gender affected the levels of blood metabolites, plasma acute phase proteins and oxidative stress biomarkers, whereby the highest concentrations of lactate ($P=0.021$), glucose ($P<0.0001$), ceruloplasmin ($P<0.0001$) and AOPP ($P<0.0001$), but the lowest GSH ($P<0.0001$) levels were recorded in stallions subjected to high loading density. Interactive effect of loading density and gender affected the occurrence of carcass bruises in slaughter horses. Furthermore, the highest percentages of severe ($P=0.0002$), large ($P<0.0001$) and circular ($P=0.0001$) carcass bruises, chiefly located on abdominal ($P=0.0056$) and thoracic ($P=0.0004$) wall, were found in stallions subjected to high loading density. In conclusion, the results of this study showed that high loading density, regardless of gender, negatively affects horse welfare during transportation. In addition, stallions were more sensitive to poor pre-slaughter conditions, while geldings were the most resistant. In order to define optimal travel density, additional research is necessary to determine the effects of different loading density on behaviour, physiology and carcass and meat quality in slaughter horses.

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Poster 10

Is observation of horses when they are outdoors adequate for detecting stereotypic individuals?

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The study's first objective was to determine if the observation of horses during turn out is sufficient in detecting stereotypic individuals. To do this, the behavioural repertoire of stable-housed horses when in the box and during turn out were compared. The second objective was to determine if living permanently outdoors (OS) resulted in less abnormal behaviour in horses as compared to those that were turn out for only four hours per day (IS). 26 mares and 13 geldings different types of breed were observed (IS: $n=19$, OS: $n=20$). All horses were observed for five hours for two days at 1-month interval. It was observed – stereotypic (SB) behaviour: crib-biting, grid biting, tongue playing, incisors sliding, weaving, box-walking, wall-kicking and aberrant (AB): licking, wood chewing, coprophagy, geophagy, pawing, aggression to humans and/or horses. Our results are in agreement with previous studies that outdoor, and may be more importantly, group keeping, is beneficial to horses, as it reduces repetitive and aberrant behaviour. Although the paddocking clearly reduces the SB, this may be also due to the lack of the substrates for performing such behaviours. When paddocks are fenced with electric fences, the horses may have even less opportunity to present crib-biting and wood-chewing. Then, preliminary and short observation of the horse outside the stable may be misleading when assessing the welfare of such a horse. Clearly, the animal is in better environmental (and often social) conditions, but this does not preclude it from presenting stereotypic behaviour when in the stable. It is concluded that, in welfare assessment protocols, the observation of horses kept for most of their time in the stable, the periods when they stay in their home boxes, should be considered. Funded by the Minister of Science under „the Regional Initiative of Excellence Program”

The influence of selected factors on the behavior of horses outside the stable during different seasons

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The influence of selected factors on the behavior of horses outside the stable during different seasons Ewa Jastrzębska and Aleksandra Pawłowska* Department of Horse Breeding and Riding, Faculty of Animal Bioengineering, University of Warmia and Mazury, Oczapowskiego 5, 10-719 Olsztyn, Poland *Presenting author: Aleksandra Pawłowska, aleksandra.pawlowska@uwm.edu.pl Horse behavior is highly diverse and influenced by multiple factors. The aim of this study was to investigate the influence of selected factors on the frequency of social and comfort behavior in horses residing in a sandy paddock and pasture during different seasons. The study included a herd of 20 horses – 14 geldings and 6 mares. The youngest was 3 years old, the oldest was 21 years old and the rest of the horses were in the 4 to 19 age range. The research involved observing the behavior of horses outside the stable, on the pasture and in the sandy paddock during four seasons. An ethogram was created to document specific social and comfort behavior. Weather conditions data for each day were collected using an anemometer. Statistically significant differences were observed in the occurrence of specific behavior between mares and geldings. The age of the horses did not affect the occurrence of specific behavior. Statistically significant correlations were found between weather conditions during the four seasons and the occurrence of social and comfort behavior. The research demonstrated that gender and weather conditions influence the occurrence of social and comfort behavior in horses. Funded by the Minister of Science under “the Regional Initiative of Excellence Program”.

Session 75

Theatre 1

Effect of starch source in pelleted diets on growth performance of pigs

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In European pig diets, wheat (W) and barley (B) are major energy suppliers. Their contrasted proportions in rapid and slowly digestible starch may alter the positive effect of pelleting on digestibility and metabolic use of energy. This study aimed to compare performance of growing pigs fed with pelleted diets, formulated for being iso-net energy (NE), either based on W, B or a balanced mixture of both cereals (so-called WB). Two batches of 224 gilts and entire males were studied between 31 and 117 kg body weight (16 pens of 7 pigs/treatment). Pigs were fed ad libitum with a 2-phase strategy. Based on chemical and nutritional (assessed with Evapig® for diets produced without any heating process) characteristics of feedstuffs, all diets were formulated to 9.40 MJ NE, 47 g crude fibre and 30 g crude fat/kg. Digestible lysine content was 0.93 and 0.85 g/MJ NE and starch content was 438 and 441 g/kg in Phase-1 and Phase-2 diets, respectively. All diets were pelleted (pellets temperature = 78±2°C). The average daily feed intake of W pigs was lower than that of B and WB pigs (2.62 vs. 2.69 kg/d on average for B and WB, P=0.03), without any difference in growth rate (1029 g/d on average for all groups, P=0.55). The resulting feed conversion ratio tended to be lower in W pigs (2.53 vs. 2.59, P=0.07). The hot carcass yield was better in W pigs (79.8 vs. 79.4%, P=0.02), but no difference was observed on carcass leanness (61.5%, on average for all groups, P=0.28). These results would agree with a greater improvement of the digestibility and/or metabolic utilisation after pelleting of W diets (compared to B), but more investigations are required to understand why results with WB were not intermediate between W and B.

Empirical validation of implementing high energy in soybean meal: soybean oil equivalence of soybean meal in diets fed to growing pigs

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The objective was to test the hypothesis that the energy value for soybean meal (SBM) fed to pigs is greater than current book values. Four diets based on corn, soy protein concentrate, and synthetic cellulose contained 0, 2, 4, or 6% soybean oil (SBO). The fifth diet contained corn, soy protein concentrate, and 12% SBM, but did not contain synthetic cellulose or SBO. A total of 120 pigs (initial weight = 24.7 kg; standard deviation = 3.06) were allotted to the six diets with two pigs per pen and 12 replicate pens per treatment. Pigs were fed experimental diets for four weeks. Average daily gain, average daily feed intake, and gain to feed ratio (G:F) were calculated for each treatment. The statistical model included diet as fixed effect and replicate and weanling group as random effects. Linear and quadratic effects of increasing SBO in diets were determined using contrast coefficients. Regression of G:F for pigs fed diets containing no SBM against the increasing levels of SBO was performed and an equation was generated. Results demonstrated that the overall G:F of pigs was increased (linear, $P < 0.05$) by increasing SBO in diets containing no SBM. From the prediction equation it was calculated that the G:F of pigs fed the diet containing 12% SBM corresponded to the inclusion of 4.84% SBO in the diet. Assuming there is 7,504 kcal net energy (NE) in SBO, this corresponds to a NE value of 363 kcal/kg from adding 12% SBM and removing 12% cellulose, which corresponds to 3,027 kcal NE per kg SBM. The hypothesis that there is more NE in SBM than indicated in current book values, therefore, was confirmed. In conclusion, the improvement in G:F obtained by adding SBM to the diets was not different from the improvement in G:F obtained by adding 4.84% SBO to the diets, which indicates that SBM provides more NE to pigs than previously thought.

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Theatre 3

Dietary fiber supplementation during gestation promotes placental angiogenesis by gut microbiota regulation of serotonin signaling pathway in sows

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The study aimed to investigate the potential mechanisms of dietary fiber improving the placental development of sows. Thirty gilts were assigned randomly to two groups, and fed a basal diet (CON group) or a basal diet supplemented with inulin and cellulose [high-fiber (HF) group] during gestation period. Sow blood and feces samples were collected on gestation day 30 for metabolomics and metagenomic analysis, respectively, and placenta samples were collected immediately after farrowing in the third parity. The results demonstrated that dietary fiber changed the serum metabolites related to tryptophan metabolism pathway, and decreased serum indole concentration. Dietary fiber supplementation increased the Firmicutes bacterium CAG.110 abundance which was negatively correlated with the serum indole concentration. Then the serum concentration of serotonin was found to be higher in the HF group compared with the CON group. Moreover, CD31 protein expression of the HF group in placenta was significantly higher than that in the CON group ($P < 0.05$). The study in vitro showed that 75 $\mu\text{mol/L}$ serotonin increased the cell viability and angiogenesis of porcine vascular endothelial cells; meanwhile, serotonin elevated the serotonin receptor 1B and PI3K/Akt pathway-related genes expression as well as the CD31 protein expression ($P < 0.05$). Our finding indicated that dietary fiber supplementation during gestation promoted placental angiogenesis by gut microbiota regulation of serotonin signaling pathway in sows.

Effects of high fibre on gas production and net energy in diets fed to group-housed pigs

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The objective was to test the null hypothesis that a high-fibre diet does not affect gas production or net energy (NE) in pigs. One diet was formulated based on corn and soybean meal with normal fibre concentration. The second diet contained 33% wheat middlings and had an elevated level of fibre. A total of 24 growing pigs (initial weight = 40.9 kg; standard deviation = 1.72) were allotted to the 2 diets using a 2-period switch back design with 6 calorimetry chambers (4 pigs/chamber) and 2 periods. Pigs had free access to feed and water. Fecal and urine materials were quantitatively collected and consumption of O₂ and production of CO₂ and CH₄ were measured for 6 d. After collection, pigs were deprived of feed for 48 h and O₂ consumption, CO₂ and CH₄ synthesis, and urine N were measured to determine fasting heat production. The statistical model included diet as the fixed effect. Results indicated that apparent total tract digestibility of gross energy and concentration of NE were reduced ($P < 0.01$) in the high-fibre diet compared with the normal-fibre diet. Respiratory quotient was greater ($P < 0.01$) if pigs were fed the normal-fibre diet than the high-fibre diet. When the gas consumption and production were corrected for feed intake, the CH₄ production was greater ($P < 0.05$) in the high-fibre diet, which is associated with greater fermentation in pigs. Total heat production was greater ($P < 0.01$) in the normal-fibre diet compared with the high-fibre diet, indicating that the metabolic activity in pigs was increased by greater energy and nutrients available from the normal-fibre diet. In conclusion, feeding pigs with the high-fiber diet reduced NE in diet because wheat middlings has lower energy digestibility and increases fermentation.

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Theatre 5

A review on the lysine requirements of weaned piglets

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Optimizing pig health and production efficiency and minimizing N excretion requires accurate knowledge of lysine requirements. This study aimed to determine standardized ileal digestible (SID) lysine requirements of weaned piglets (5-30 kg) based on a literature review using meta-analytical approaches. The literature review yielded 344 studies that were screened for title and abstract. In total, 41 experiments met the inclusion criteria, resulting in a dataset of 206 treatment groups. Linear, quadratic and linear-plateau models were used to obtain insight in the effect of SID lysine content on average daily gain and feed efficiency for the combined dataset and for individual experiments. For the combined dataset, both the dietary lysine content and the response criteria were standardized. Regression analysis showed a predominantly linear increase in average daily gain and feed efficiency in response to increasing lysine levels across both the combined dataset and individual experiments. Breakpoint estimation from the linear-plateau models was inconclusive, indicating that the optimal SID lysine requirement to maximize piglet growth performance likely exceeded the upper lysine levels tested in most studies, surpassing 1.3 g SID lysine per MJ net energy. This review indicates a high value for the dietary lysine content required to maximize growth performance. Results may also suggest that piglet feed formulation should focus on an optimal dietary SID lysine to crude protein ratio, rather than SID lysine per kg of diet or unit of net energy.

Estimating the efficiency of amino acid use using chemical composition data from growing pigs

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In recent decades, phenotypic, anatomical and chemical changes have been observed in pigs. As these developments have implications for pig nutrition, it is important to regularly update the databases used to estimate nutritional requirements. In this context, the aim of this study was to estimate amino acid (AA) use efficiencies using data on pig body composition from experimental trials conducted over the last 10 years. The database thus created consisted of data of growing pigs of three sexes (i.e. castrated males, whole males and females) fed according to Swiss recommendations or with a limited protein diet, and formulated according to the ideal AA profile or not. Slaughter data for these pigs and analysis of their AA composition were also available. The individual and daily intake of the pigs and the net energy, protein, and AA composition of the feed and their standardised ileal digestibility were also known. Use efficiency of methionine + cysteine and leucine differed significantly as a function of live weight ($P < 0.05$), but only tended to do so for arginine and histidine ($P < 0.07$). The use efficiency of methionine + cysteine also varied as a function of sex ($P < 0.05$), being lower for females than for whole males. Finally, the use efficiency of isoleucine, leucine, valine and phenylalanine + tyrosine was significantly higher when the diets distributed were not formulated according to the ideal AA profile ($P < 0.05$). This study updated the data on the use efficiency of many AAs by growing pigs and identified some factors that influence these use efficiencies. These data could help estimate the AA requirements of pigs more accurately.

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Theatre 7

Optimal protein concentration in diets for multiparous sows during the transition period

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Little is known about protein requirements of sows during the transition period. We aimed to determine the dietary protein required in transition diets for multiparous sows to optimize their farrowing process, colostrum production, and subsequent lactation performance. Forty-eight multiparous sows were allotted to one of six isoenergetic diets with increasing concentrations of dietary protein (expressed as standardized ileal digestible [SID] Lys: 3.99 to 8.57 g SID Lys/kg) from day 108 of gestation until 24 h after farrowing onset, and were fed 3.8 kg/d. Body weight and backfat depth were recorded, and blood was sampled on day 108 of gestation and weekly in lactation for analysis of metabolites. On day 115 of gestation, urine and feces were collected for nitrogen balance and analysis of urine metabolites. Numbers of liveborn and stillborn piglets were recorded, and piglets were weighed from birth until weaning to estimate sow colostrum and milk yields. Nitrogen utilization was maximized at 6.06 g SID Lys/kg of feed ($P < 0.01$) and the ratio of urea to creatinine in urine remained stable until 6.08 g SID Lys/kg, after which it increased linearly ($P < 0.01$). Sow body weight, backfat, plasma metabolites and colostrum yield were unaffected by treatment, but stillbirths increased linearly with increasing SID Lys ($P < 0.001$). A dietary concentration of 5.79 g SID Lys/kg during transition optimized milk production, with an average of 13.5 kg/d ($P = 0.04$). In conclusion, the transition diet of multiparous sows should contain 5.79 g SID Lys/kg when fed at 3.8 kg/d, for a total SID Lys intake of 22 g/d.

Evaluation of the soybean replacement as protein source with Mediterranean legumes in the performance of growing pigs

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The aim of this study was to evaluate the effect of soybean replacement as protein source by local legumes in terms of performance of growing pigs. Pea, lentil and chickpea were selected to replace 50 or 100% of protein from soybean in the diet, making up six groups (pea100, pea50, lentil100, lentil50, chickpea100, chickpea50) that were compared with a negative control (NC, soybean as main protein source). A total of 252 pigs (20.3±1.34 kg) were allocated to 7 experimental treatments (6 replicates/treatment; 6 pigs/replicate). Diets were formulated to be isonutritive (13.1 MJ/kg ME and 16.2% CP). Pigs were weighed on day 0 and 28 of trial. Average daily gain (ADG), average daily feed intake (ADFI) and gain feed ratio (GFR) were calculated. Fecal samples were taken directly from the rectum on days 24-28 to determine apparent fecal digestibility of dry matter, organic matter and protein. Statistical analysis was conducted by ANOVA. Contrast analyses were conducted between NC and each legume. The highest ADG and ADFI were obtained in pigs fed the pea and NC diets. Pigs fed the lentil diets had the lowest ADG and pigs fed chickpea were intermediate ($P<0.05$). Pigs fed pea diets showed the highest feed efficiency ($P<0.05$) and pigs fed the lentil diets had the lowest GFR. Pigs fed the pea100 diet showed the highest nutrient digestibility values, while pigs fed the chickpea diets showed the lowest ones. To sum up, peas seem to be the best alternative to replace soybean as protein source in terms of performance and nutrient digestibility. Funded by European Union's Horizon Europe Research and Innovation Program, Grant agreement No. 01059609 (Re-Livestock).

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Theatre 9

Sustainable protein sources field peas and blue lupines replace soy protein in piglet diets

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To study if peas or lupines can replace soy in pig diets three piglet trials were conducted. In the first trial (n=48), the nutrient digestibility was measured for a diet with 18% soy bean meal or 19% sweet lupines. In the 2nd trial (n=46 piglets), the nutrient digestibility was measured for a diet based on soy protein or a diet in which part of the soy was replaced by 7.5% peas in the 2nd week or 15% peas in the 4th week. In the third trial (n=393), the performance was measured of pigs either fed a diet based on soy or 7.5% and 15% peas or 5% and 10% sweet lupines in weaner and rearing diet respectively. In the first digestibility trial there was no significant difference in nitrogen absorption and nitrogen retention for the piglets fed diets with soy or lupines in the 2nd week or in the 5th week post weaning. Also the fat absorption was not significantly different between piglets fed the diet with soy or lupines in the 2nd or 5th week post weaning. In the 2nd digestibility trial there was no significant difference in nitrogen absorption and nitrogen retention between the diets in the 2nd week (79.4% vs 80.0% and 70.1% vs 69.6%) or in the 4th week post weaning (78.8% vs 77.5% and 69.4% and 68.6%). The fat absorption for piglets was not significantly different in the 2nd week (66.6% vs 66.9%). However, in the 4th week post weaning, the fat absorption was higher ($p<0.001$) for the piglets fed the diet with peas (68.5% vs 72.8%). In the performance trial the feed intake, weight gain and feed conversion ratio did not differ significantly between the treatment groups. These trials show that sustainable protein sources peas and lupines can replace soy in post weaning piglet diets without compromising performance.

Metabolism of phytoestrogens in red clover and effects on sow reproduction

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Red clover (*Trifolium pratense*) contains phytoestrogens (PE), such as formononetin, biochanin A, daidzein and genistein. During digestion, PE are metabolised to equol, which could have negative effects on reproduction. However, studies on metabolism and effects on sow reproduction is limited. In a first study, 8 gestating sows were fed red clover silage (RC) for 4 weeks and feed, faeces and urine were analysed for PE content. A second study included 23 sows, 11 fed PE rich red clover silage (RCS) and 12 fed white clover (*Trifolium repens* L.) silage with no PE (WCS) during two reproduction cycles (r1, r2). Heat signs, teat characterization and udder filling were registered at day 30, 60 and 90 after insemination in r1 and r2, and faeces, urine and blood were analysed for PE at day 0, 30 and 60 in r1. Samples were processed with microwave-assisted and solid phase extraction and analysed by UHPLC. PE and quantitative reproduction data were statistically analysed with mixed model and repeated measures. Most PE in faeces and urine in study 1 differed over time, but with no consistent pattern. Faecal equol increased gradually from w1 to w4 ($P=0.011$) and in urine from w1 to w3 and 4, where it reached a plateau value ($P=0.006$). The unconjugated forms of PE and equol in urine were low or under detection level. In study 2, PE was analysed, and preliminary results showed no effect of RCS and WCS silage or reproduction cycle on reproduction traits ($P\geq 0.05$) and all sows had normal udder filling. Sows fed red clover silage metabolize PE to equol. Most PE in urine were found in the conjugated form and equol reached a plateau value of 208.8 mg/mL.

Session 75

Poster 11

A comparison of Devamine and traditional protein sources on the post-prandial release of blood metabolites in weaned pigs

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Protein sources differ in digestion and absorption kinetics, influencing pigs' amino acid balance, protein efficiency and ammonia emissions. This research delves into the effects of protein sources on blood metabolites in weaned pigs. Ninety-six piglets ($14.5\text{kg} \pm 0.35$) were not offered feed for 12 hours, then fed a diet containing either vegetable protein (VP), synthetic amino acids (SYN), specialised protein product (Devamine), or a combination of sources (COM). Blood samples were collected post-feeding and analysed using GC-MS, with ANOVA for statistical analysis. The results revealed no significant differences in total plasma amino acids, urea, or the amount of urea produced per nmol of amino acids absorbed at 0 minutes. At 60 minutes, the combination diet significantly increased total plasma amino acids compared to vegetable protein and Devamine (824 vs 241 vs 239 ± 146.2 nmol/ml, $P<0.05$). While the combination diet tended to increase urea (6.02 vs 2.52 vs 2.99 ± 0.88 umol/ml, $P<0.1$), there was no increase in the amount of urea produced per nmol of amino acid absorbed. The Devamine diet at 360 minutes resulted in a significant increase in total plasma amino acids compared to all other treatments (1113 vs 273 (VP) vs 193 (SYN) vs 408 (COM) ± 222.0 nmol/ml, $P<0.05$). Despite the significant increase in plasma amino acids, there were no significant increases in urea, and a tendency for Devamine to reduce the amount of urea produced per nmol of AA absorbed compared to synthetic AA (0.015 vs 0.023 ± 0.005 umol/nmol, $P<0.1$). As urea is a by-product of AA catabolism, the lack of increase in urea when utilising Devamine in the diet suggests that catabolism is not increased despite the increase in plasma AA. The study concludes that the inclusion of Devamine can aid protein efficiency by increasing plasma AA levels without an increase in catabolism. In contrast, synthetic amino acids appear to increase catabolism, which would reduce protein efficiency.

Seaweed formulation enhances sow performance through reduced stillborn piglets and improved vitality
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A thriving piglet at birth is vital for survival and growth, which is strongly influenced by farrowing length. Weakness and potential death are especially a risk for later born piglets due to asphyxia when farrowing takes too long. Reducing farrowing time has a reported positive effect on sow and litter performance. Data suggests this can be achieved by a better energy availability, specifically through glucose uptake by muscle cells. Multiple research studies have investigated the potential of a seaweed formulation (PS) with anti-inflammatory bio-actives and marine minerals to improve farrowing time and sow performance. The objective of this study was to investigate the robustness of the efficacy of PS on sow performance in a large sow unit with 7,200 sows in Iowa, USA. PS was introduced at the farm in both gestating and lactating diets at 0.4% inclusion, replacing equal amounts of limestone. An 11-week control period prior to the start of PS was followed by an 8-week transition period and subsequently a 22-week PS observation period. Farrowing performance measures were; number of total born (TB), liveborn (LB), and stillborn (SB) piglets and pre-weaning mortality (PWM). Data was analysed by Excel-Stat (16.7, 2022.4.1) using Tukey's adjustment. The PS inclusion resulted in better farrowing performance, with an increase in TB and LB piglets compared to the control period (TB: 15.6 vs. 16.3, SEM 0.10; P<0.05 and LB: 13.8 vs. 14.7 piglets; SEM 0.09; P<0.05). The SB incidence reduced in the PS period compared to the control period (8.25% vs. 6.86%; SEM 0.339%; P<0.05). PWM was also reduced in sows fed PS to 14.7% while this was 18.6% in the control period (SEM 0.50%; P<0.05). In conclusion, PS robustly improves sow performance on farm, validating previous research study findings on improved farrowing time leading to better piglet survival.

The Microbial Profiling of the Boschveld Chickens Fed Sorghum-Based Diets
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This study aimed to evaluate the effects of feeding sorghum-based diets on the gut microbes of Boschveld indigenous chickens. A total of 420 unsexed day-old chicks were assigned in a completely randomized design with 3 sorghum varieties x 5 inclusion levels x 4 replicate with 7 chicks per replicate for 90 days. Two chickens per replicate were slaughtered at days 60 and 90, and caeca were collected and stored in tubes filled with 70% ethanol and immediately kept in ice. Collected samples were processed using the 16s rRNA metagenomics sequencing on Illumina's MiSeq platform. The resulting amplicons were purified, end-repaired and Illumina-specific adapter sequences were ligated to each amplicon (NEBNext Ultra II DNA library prep kit). Analysis was done using in-house python scripts version 3.6.1. kronaTools and the Rstudio software following phyloseq package R version 3.5.0. The bacterial communities at alpha diversity indices at observed, Shannon and Simpson were more abundant at day 90 than day 60. The bacteria communities dominated at phylum level were Firmicutes, Proteobacteria, Bacteroidia, while Lactobacillus – Avarius, Comamoducaae and Methylobacterium were dominating at genus levels. This study shed an understanding of the bacterial diversity influenced by feeding different diets dominating chickens' gut microbiome.

Spirulina as alternative protein source in diets for gestating and lactating sows – effects on litter characteristics and piglet growth

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The ban on synthetic amino acids in organic pork production and the general aim to reduce the critically discussed soybean meal in pig feeding pose challenges in providing adequate essential amino acids to sows. Excess dietary protein, often from lower-quality sources, is common. *Arthrospira platensis* (spirulina), rich in protein and essential amino acids, emerges as an alternative. In our study, 9 multiparous sows (205 ± 10.2 kg body weight) were divided in two groups and fed one of two diets; either a commercial control diet (CON; n=5) or an experimental diet in which soybean meal and rapeseed cake were (partially) replaced by spirulina (SPI; n=4). In the gestation diet, soybean meal was replaced completely, and rapeseed cake was reduced by 52%, while in the lactation diet, only soybean meal was reduced by 42%. Gestation and lactation diets supplied about 133 g and 174 g, respectively, crude protein per kg as fed. Results showed no significant differences in litter size, weight, or individual piglet birth weight. However, SPI piglets were heavier at one week compared to CON piglets (3.1±0.19 vs. 2.5±0.16 kg; p=0.014). Also at weaning, body weight was higher in SPI compared to CON piglets (9.0±0.31 vs. 8.1±0.27 kg; p=0.026). Still, average daily gain did not differ significantly from birth to weaning. The study suggests replacing soybean meal and rapeseed cake with spirulina in sow diets does not hinder offspring growth and may lead to heavier piglets at weaning.

Session 75

Poster 15

Effect of consumption of A1 and A2 milk, biofortified or not, on weight gain and carcass yield of piglets

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To evaluate the effect of ingesting biofortified or non-biofortified milk from cows with genotypes A1A1 and A2A2 for the beta-casein gene on performance in the growth phase and carcass yield of Landrace Large White hybrid piglets. Forty piglets, aged 28 days with an initial average body weight of approximately 9 ± 1.0 kg, were housed in collective pens (4 piglets per pen) and subjected to CONTROL treatments (solid diet + dehydrated lactose); A1BIO (solid diet + biofortified A1 milk); A1NB (solid diet + non-biofortified A1 milk); A2BIO (solid diet + biofortified A2 milk) and; A2NB (solid diet + non-biofortified A2 milk). The piglets were weighed at birth, weaning (28d) and then at 42, 58, 75, 84 days (slaughter) and carcass yield was calculated. All analyzes were carried out in SAS version 9.4 (SAS Institute Inc.) using a completely randomized block design in a 2 × 2 + 1 factorial arrangement, in which genotypes were considered as factor 1, biofortification as factor 2 and the control as an additional factor. For the variable weight at 58 days, a significant interaction effect was observed between the factors (p-value = 0.01) in which A1NB (18.7) and A2BIO (19.9) presented the highest means. For the variable weight at 84 days, a significant interaction effect was also observed between the factors (p-value = 0.04) in which A1NB (37.6) presented the highest average. Piglets subjected to A1NB and A2BIO treatments showed the highest weights at 58 days and 84 days, indicating that feeding increased the animals' growth potential.

Golden mussels meal with varying particle sizes in broiler chicken diets

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This study aimed to assess the impact of golden mussel meal (GMM), replacing limestone, with varying particle sizes in broiler chicken diets, on intestinal transporters gene expression. A total of 900 male Cobb 500® broiler chicks were allocated in a completely randomized experimental design with 5 treatments, 10 replicate pens, and 18 birds per experimental unit (EU) from 1 to 40 days of age. Golden mussels were collected from the Itaipu Hydroelectric Power Plant Reservoir (Porto Mendes, Paraná). Treatments comprised a control diet (calcite limestone), GMM 299 µm, GMM 375 µm, GMM 436 µm, and GMM 551 µm. Jejunum samples from 1 bird per EU (n= 50) at 40 days were collected and stored in RNA Later solution for evaluating gene expression of Calbindin-D28k (CalbD28K), cell receptors sensitive to Ca (CaSR), Ca and Na carriers (NCX1), and Ca Plasma Membrane (PMCa). RNA was extracted using a commercial kit QIAzol (Qiagen®), with the β-actin gene serving as an endogenous control. Treatment effects were verified using ANOVA, and significant results analyzed using polynomial regression. The Dunnett test was employed to compare treatments with the control (limestone). A significance level of α=0.05 was used for all tests. SAS University Edition software was used for statistical analysis. No differences were observed in gene expression of intestinal transporters in the jejunum (P>0.05). The replacement of calcium by GMM, regardless of particle size, showed no adverse effects on CalbD28K, CaSR, NCX1, and PMCa gene expression, thus not affecting calcium absorption in the jejunum.

Session 75

Poster 17

Digestibility assessment of various protein sources in cat feed

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Cats are obligate carnivores. Their digestive systems are optimized to digest protein and fat quickly. This study assessed the digestibility of major protein sources frequently used in cat feed, focusing on beef, chicken, dried pollock, duck, and salmon. The selection of these sources was based on a survey of 55 products from the top six brands in South Korea. From the analysis of total protein content and amino acids composition in the processed materials and test feeds with a single protein source, the protein content averaged $61.6 \pm 4.76\%$ and $25.72 \pm 1.4\%$ in the processed materials and the testing feeds. In addition, glutamate was the predominant amino acid in both processed materials (8.17%) and testing feeds (3.27%), while tryptophan was the least abundant (0.45% in processed materials, 0.12% in test feeds). When the digestibility of each protein source was evaluated in 10 cats (the ratio of male to female, 1:1; age range from 1 year to 5 years) using a test feed based on a single protein source, the protein digestibility of beef, chicken, dried pollock, duck, and salmon were 73.81%, 82.05%, 71.81%, 80.64%, and 75.01% respectively. From the digestibility of each amino acid, cystine and tryptophan were the highest in chicken-based feed, while all other amino acids including alanine, arginine, aspartic, glutamate, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tyrosine, and valine were the highest in duck-based feed. These results could contribute to the development of feed formula for cats and the improvement of animal welfare.

Bioactive compound research in ruminant nutrition: From Reductionism to Holism

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Bioactive compounds are chemical compounds produced by plants for their defense against microbial and insect attacks. Since the EU-wide ban on the use of antibiotics as growth promoters in animal feed in 2006, there has been growing research on phytochemicals for modulating gut health and improving the productivity of livestock species. In ruminant research, this is further fueled by the possible effects of bioactive compounds on decreasing methanogenesis. Three groups of bioactive compounds heavily researched are saponins, phenolic compounds (tannins and non-tannins), and essential oils from different botanical origins. There are commercial products, which are usually blends of plant extracts and bioactive compounds, sold as feed additives for livestock species. This underlines the impacts of bioactive compounds on the feed system and consequently our foods. Novel sources and other groups of bioactive compounds have been explored. Despite the big body of data, the implication and success of feed additives are still challenged by the fact that the modes of action of phytochemical additives are not strictly governed by the classification of the bioactive compounds. Different factors influence the property, biochemistry, and thus the effect and efficiency of a compound under a given environment, not to mention the bioavailability and pharmacokinetics that have been rarely considered in livestock research. Stemmed from the collective work of the author as well as from literature, the present work will summarize the existing knowledge and challenge the future of bioactive compound research in ruminants, including target use of phytochemicals during stressed rumen conditions, novel sources of bioactive compounds, a whole-plant concept, and effects beyond rumen.

Session 76

Theatre 2

The impact of probiotic live yeast in a barley grain-based diet on rumen microbiome and histology of artificially reared lambs

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Forty-two Chios lambs were allocated into two groups: Control (CON) and Probiotic group (PROB) and were artificially reared until the 45th day of age. Both groups were fed alfalfa hay and a barley grain-based concentrate mix ad libitum while PROB lambs were also fed 100 g of libitum including 0.1 g of *Saccharomyces cerevisiae* CNCM I-1077 (10¹⁰ CFU/g). Rumen digesta was collected on the 45th and 100th day from 10 lambs per treatment using a stomach tube while on the 106th day, rumen tissue was collected at the slaughterhouse. Although α - and β -diversity remained unchanged across dietary treatments, differences were observed in microbial composition over time (45d vs. 100d). PROB diet caused an increase of *Butyrivibrio*, *Anaerovibrio*, *Pseudobutyryvibrio*, *Sphaerochaeta*, and *Ruminobacter* and a decrease of *Clostridia* UCG-014 and *Acidaminococcus* at 100 days. Rumen histology observations indicated that PROB lambs' mucosa histology outweighed that of the CON lambs as regards the appearance and distribution of the extracellular matrix components (collagen and reticular fibres), the prekeratin and keratin contents and finally the appearance of the epithelium, which was much less keratinized. In conclusion, the formulation of a barley grain-based concentrate mix might induce a low-grade rumen dysbiosis that in the case of PROB-fed lambs was more attenuated.

Rutin and Quercetin Biotransformation in Ruminants Revealed by Molecular Networking

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Quercetin and its glycoside, rutin, are flavonoids that are widely used as robust antioxidant and anti-inflammatory agents for improving animal health. They are metabolized into various phenolic compounds, which may be more potent than their original forms. However, the biotransformation pathways for quercetin and rutin in ruminants have not yet been fully elucidated. To identify this pathway, we used a molecular networking approach to determine the metabolite formation of ingested rutin and quercetin. Five Shiba crossbreed goats were used in a cross-over design and categorized in three treatment groups: rutin, quercetin, and control. Rumen fluid was collected at 0, 1, 4, 6, 8, and 22 h, while blood was collected at 0, 4, and 8 h after administration and were subjected to LC-MS/MS. A biomarker test was conducted to evaluate the performance of metabolite. Univariate data were analyzed using ANOVA. According to findings from molecular networking, rumen microorganisms break down rutin to produce quercetin, which is then further broken down into phloroglucinol (PG) and 3,4-dihydroxyphenyl acetic acid (3,4-DHPAA), respectively. Protocatechuic acid (PCA), 3-(3-hydroxyphenyl)propanoic acid (3-HPPA), and 4-methylcatechol (4-MC) were the products of the biotransformation of 3,4-DHPAA. The main phenolic compound in blood was 4-MC sulfate, a conjugate of 4-MC, produced via sulfation. PG was metabolized to 3,5-dihydroxycyclohexan-1-one (3,5-DHDC) and remained undetectable in the blood. The major flavonol and quercetin conjugate found in blood was isorhamnetin glucuronide-sulfate. In conclusion, quercetin is mainly metabolized to 4-MC sulfate and isorhamnetin glucuronide sulfate in ruminants.

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Theatre 4

Preliminary study on some innovative rumen-bypass micro-encapsulation techniques for essential oils

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The aim of this trial was to provide novel technologies for the production of rumen by-pass essential oils to be released in the abomasum. For this purpose, 99.98% of pure limonene (Lim) was microencapsulated. Microparticles were produced using the prilling/vibration technique. An emulsion with Lim oil and alginate was combined with Eudragit EPO 30% (w/w) or Edraguard® 30% (w/w) for the production of the 3 experimental formulations (F). In vitro digestion (IVD) trial for each F was performed on rumen fluid into quadruplicate. IVD was conducted for 6, 12, 18 and 24h using the Daisy II incubator system. In each jar, 14 bags were introduced, each containing 0.5 g of microcapsules. The content of the bag was weighed to calculate the disappearance (w/w), and rumen fluid was sampled for Lim quantification. The concentration of Lim was carried out using SPME/GC-MS. The F1 and F2 had a disappearance of over 50% after 6h of digestion ($P < 0.01$), increasing to more than 80% and 70%, respectively after 24h ($P < 0.01$), while, in F3 no significant disappearance ($P > 0.05$) was observed, ranging between 0.5% (6h) and 1.3% (24h). Lim content ranged from 1472 µg/L at 6h to 1959 µg/L at 24h for F1, from 1021 µg/L at 6h to 1488 µg/L at 24h for F2, and from 2 µg/L at 6h to 9 µg/L at 24h for F3, respectively. Two out of the three formulations (F1 and F2) showed low resistance in the rumen environment. In contrast, the use of Edraguard® in core-shell exhibited a very high resistance up to 24 hours, laying the groundwork for future research on the microencapsulation of substances characterized by rumen-bypass.

How can drinking water may be used to increase milk yield?

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The aim of this study was to measure the effect of drinking water salinity and hardness (Ca + Mg) on lactating cow's performance. Genetic selections, optimizing feed, inseminations, and housing conditions are among the major strategies aimed to improve dairy industry production end profit, with numerous studies focusing on those topics and more. Absent from the list is a strategy to exploit the ability to control drinking water minerals composition. The common practice is to ensure unlimited access to fresh water and as long that the water are not too loaded with dissolved solids (<1000 ppm) it considered safe to drink. However, water is one of the most consumed nutrient required for living and lactating cows consume it in a large volume diverting it to milk, urine, feces, and evaporation. After construction of experimental systems capable to control and measure individual drinking, and executing two independent trials (4 by 4 Latin square), we found that lactating cows (Israeli Holstein) drink ~142 l in average of 8 drinking events / d from few and up to 83 l in one event. Cows drank 120 to 160 l / d, positively related to feed intake ($R_2 = 0.66$, $p < 0.01$). Results of first trial, conducted in summer 2022, examined the effect of drinking water salinity, by adding NaCl to create water EC of 400, 600, 800, and 1000 $\mu\text{S}/\text{cm}$, on lactating cows performance, showed increased milk yield of up to 2 kg / d when water salinity increased. In our second trial, conducted in summer 2023, we found that increasing drinking water hardness from 80 to 160, 240, or 360 ppm (CaCO₃ equivalent) by adding 0, 46, 92, or 138 mg / l CaCl₂ + MgSO₄ (Ca: Mg ratio of 2:1) increased milk yield by up to 2.2 kg / d. Those results imply on the potential of using drinking water to improve production and require more research in the topic.

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Theatre 6

Water intake and productivity responses of lactating dairy cows offered saline drinking water

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Water scarcity in certain Swedish regions necessitates securing potable water for cattle and exploring alternative sources. The brackish Baltic Sea (2-8 g NaCl/L) is commonly used as a water source for grazing beef cattle but the potential as a water source for high-yielding dairy cows is not known. This experiment investigated the effects of saline drinking water (8 g NaCl/L) on lactating dairy cows. Eighteen cows in mid-lactation of the Swedish Red and Holstein breeds were used in an unbalanced Latin-square design. Cows received three treatments: control (CON; < 1g NaCl/L), low salinity (LOW; 4 g NaCl /L) and high salinity (HIGH; 8 g NaCl/L) water for 14 days per treatment. Daily water intake and milk yield were registered and were analysed using RStudio. There was a significant effect of treatment on milk yield ($p = 0.047$) and water intake ($p < 0.001$). Milk yield averaged 33.4 ± 1.13 kg/d (CON), 34.1 ± 1.13 kg/d (LOW) and 33.9 ± 1.13 kg/d (HIGH), while water intake averaged 95.4 ± 4.26 kg/d (CON), 113.1 ± 4.26 kg/d (LOW) and 147.2 ± 4.26 kg/d (HIGH). The findings indicate the potential for utilizing Baltic Sea water to high-yielding dairy cows without compromising milk production.

Does Exhalomics Reveal the Dynamics of Rumen Fluid Metabolites in Dairy Cows?

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This study aimed to explore the potential of exhalomics (EX) vs. rumen fluid (RF) analysis to understand fermentation dynamics across a feeding day, using a switchback design over three 9-d periods (7-d adaptation + 2-d sampling). Four rumen-cannulated Swiss Brown cows were enrolled in (A) low-starch [6.31% of DM (dry matter)] and (B) high-starch (16.2% of DM) diets in an ABA/BAB sequence. Cows were fed 1×/d at 0830 h. Exhalome (with the GreenFeed® System), and RF samples were collected 8× to represent every 3h of a day and analyzed using secondary electrospray ionization-mass spectrometry and liquid chromatography-mass spectrometry, respectively. Raw features were scaled (Pareto) and analyzed in R and MetaboAnalyst. In total, 1,096 RF and 1,694 EX features were detected. Metabolomic data at 3h intervals separated HS vs. LS diets in both EX and RF profiles using sparse PLS-DA ($P < 0.05$; EX: 83.3%, RF: 91.8% accuracy). Diurnal metabolite alterations (direction: HS-to-LS) showed the most significant changes 2h after feeding in RF (37 up- and 39 downregulated metabolites; $P < 0.05$), suggesting a high fermentation rate which gradually diminished and stabilized before the next feeding. In contrast, EX-profile showed a lower change magnitude starting 2h post-feeding and upregulated (24 metabolites; $P < 0.05$) till noon. In conclusion, exhalomics offers a non-invasive complementary for assessing fermentation dynamics, effectively capturing metabolic shifts.

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Theatre 8

Is tributyrin a good replacement for sodium butyrate to stimulate epithelial growth in sheep?

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High concentrate intake and accompanying increased ruminal butyrate production are known of their proliferative impact on the ruminal epithelium. To further increase butyrate concentration in a ruminal digesta, various sources of butyrate can be used. The aim of the study was to compare two sources of dietary butyrate: commonly used sodium butyrate (SB) and tributyrin (TB) as an alternative to SB, when those were supplemented in a high-concentrate diet of growing rams. Thirty-two rams (30.6 ± 2.5 kg; 11-14 months of age) were allocated into 4 treatments and fed ad libitum diets: 1) low (22.5%; L) or 2) high (60%; H) inclusion of concentrate in diet dry matter (DM); 3) H with inclusion of SB (3.2% of diet DM; H+SB); and 4) H with inclusion of TB (2.93% of diet DM; H+TB). After three weeks, the rams were slaughtered, short-chain fatty acids concentration in the rumen fluid was analyzed and histological measurements of the ruminal epithelium were performed. The pre-planned contrasts were used for data analysis: L vs. H, H+SB and H+TB; H vs. H+SB and H vs. H+TB. Both SB and TB increased butyrate concentration ($P < 0.01$) in a rumen fluid. When compared to H treatment, H+SB resulted in increased living strata in atrium ruminis ($P = 0.03$) and increased epithelium and living strata thickness in ventral rumen ($P \leq 0.02$), while the H+TB had no impact on ruminal epithelium growth ($P \geq 0.12$). The results show that SB affects ruminal epithelium while TB does not. Therefore, TB should not be used as an alternative to SB when the target is to stimulate ruminal epithelium growth.

Identification of natural methane inhibitor based on molecular docking techniques and evaluation its effect on rumen fermentation and prokaryotic communities in vitro

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Inhibition of methyl-coenzyme M reductase can suppress the activity of ruminal methanogens, thereby reducing enteric methane emissions of ruminants. However, developing specific and environmentally friendly inhibitors is a challenging endeavor. To identify a natural and effective methane inhibitor that specifically targets methyl-coenzyme M reductase, molecular docking technology was employed to screen a library of phytochemical compounds. The screening results showed that rosmarinic acid (RA) can traverse a narrow channel and bind to the active sites of methyl-coenzyme M reductase, with a calculated binding free energy of -9.355 kcal/mol. Furthermore, the effects of RA supplementation on methane production, rumen fermentation, and the microorganism's community in dairy cows were investigated through in vitro rumen fermentation simulations. Compared with the control, supplementing RA decreased methane production ($P < 0.001$). In addition, RA supplementation increased the molar proportion of acetate ($P < 0.001$) and propionate ($P = 0.004$), whereas the sum of acetate and butyrate divided by propionate was decreased ($P < 0.001$). For bacteria, supplementation of RA decreased the relative abundance of Rikenellaceae RC9 gut group, Christensenellaceae R7 group, Candidatus Saccharimonas, Desulfovibrio, and Lachnospiraceae FE2018 group, and increased that of DNF00809 (a genus from Eggerthellaceae) and Denitrobacterium ($P < 0.05$). For archaea, the relative abundance of Methanobrevibacter decreased ($P < 0.001$), while that of Methanosphaera increased with the RA supplementation ($P < 0.001$). These findings suggested that RA exhibits potential as a novel natural additive for inhibiting ruminal methane production.

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Theatre 10

Supplemental rumen-protected methionine and lysine increase energy-corrected milk production by early postpartum Holstein cows

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Our hypothesis was that supplemental rumen-protected methionine (RPMet) and lysine (RPLys) would increase milk production during the early postpartum period. Primiparous (n=40) and multiparous (n=48) Holstein cows were blocked by calving date and lactation number and within blocks assigned randomly to 1 of 4 treatments beginning at calving. Treatments were: 1) Control (CON), basal diet AA supply without RPAA, 45 g/d of Met and 137 g/d of Lys (3.06 Lys:Met), 2) MET, supplemental RPMet, 53 g/d Met and 137 g/d Lys (2.61 Lys:Met), 3) ML, supplemental RPMet and RPLys, 53 g/d Met and 162 g/d Lys (3.04 Lys:Met), and 4) HML, high supplemental RPMet plus RPLys, 62 g/d Met and 162 g/d Lys (2.61 Lys:Met). Cows were on experiment from 24 d before expected calving and treatments continued from calving until 63 d post-calving. A mixed model with treatment, parity, time, and all interactions as fixed effects and block as a random effect was used to analyze the data. Dry matter intake tended ($P = 0.08$) to be increased by ML compared with CON (17.4, 18.4, 19.7, and 18.4 kg/d for CON, MET, ML, and HML, respectively). Milk yield tended ($P=0.06$) to be greater for ML than for CON (36.6, 38.3, 40.4, 36.6 kg/d) and energy-corrected milk was greater ($P=0.03$) for ML than for CON (34.9, 37.2, 39.6, 35.6 kg/d). Yields of milk fat and protein followed a similar pattern, but percentages of fat and protein did not differ among diets. Body weight and body condition score did not differ. Supplemental RPMet and RPLys increased early postpartum milk production, but further increasing RPMet negated the gains.

Dietary guidelines for Zn, Cu, Mn, and Fe in bovines, by integrating net requirements, native dietary occurrence, and homeostatic regulation boundaries

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Current Zn, Cu, and Mn supplementation practices exceed conservative reference guidelines. These are justified by a perceived uncertainty in basal supply and nutritional availability for these micronutrients, based on poorly understood nutritional antagonisms. Excessive supplementation is however not without risk, as supply can overwhelm homeostatic regulation mechanisms leading to excessive nutrient retention, resulting in chronic health risks. Nutritional supplementation should not only consider the lower boundaries of deficiency, but also the upper boundaries of tolerance to excess. Here we describe a novel risk-based approach to define supplemental levels for trace metal adequacy for bovines. Theoretical maximum apparent absorption efficiencies are proposed to define lower supply boundaries, whereas upper boundaries were defined as the dietary levels above which apparent absorption exceeds net requirements, thus indicating homeostatic failure. Data from literature were used to describe the probability distribution of net requirements. Similarly, the probability distribution of trace metal supply was based on variability in dry-matter intake, dietary formulation, and native metal concentration of feedstuffs. For Fe, all scenarios before supplementation met minimum needs, and most exceeded the tolerated boundary (98.9% of lactating diets, 76.4% of growing diets, and 43.0% of dry diets). For dry cows, Zn and Mn needs were met by all native diets, whereas dietary Cu supply was deemed insufficient in 30.2% of cases. During lactation, 40.7% and 2.9% of the possible scenarios were below the lower boundaries for Zn and Cu, respectively. Interestingly, all scenarios met minimum needs for Mn, and 10.8% exceeded upper tolerance boundaries. For growing heifers, 3.0%, 1.9% and 0.5% of simulated scenario, before supplementation, seemed insufficient in Cu, Mn, and Zn. Upon these results, ranges of adequate supplemental levels are proposed for dry, lactating, and growing bovines based on risks, for failure to meet sufficiency and, to exceed tolerance. Finally, these novel reference levels were evaluated against a dataset including diet formulations, analyzed trace metals in feedstuffs, and animal lactation performance.

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Theatre 12

uNDF240 and fiber digestibility as important parameters for balancing diets: comparison of wheat hay vs. wheat straw in high producing dairy cows

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Understanding the filling effect of forages on high-producing dairy cows' diets is essential for an accurate formulation. A good way to access that is through the *in vitro* undigested fraction of aNDFom at 240 hours (uNDF240), which intake goes from 0.28 to 0.48% of BW. 8 multiparous Holstein cows were used in a Latin Square design (14d adapt., 7d exp.) to investigate the effects of diets varying in wheat hay (WH) and wheat straw (WS) inclusion, both pre-chopped. These forages have the same botanic origin but a different rate of fiber degradability (52 vs. 77% aNDFom, 10 vs. 22% uNDF240, and 65 vs. 52% NDFD30, respectively). The 4 diets had a fixed amount of alfalfa hay (6.4 kg DM) and increasing amounts of WS (0, 0.9, 1.8, and 3.6 kg DM), while decreasing WH (7.5, 5.4, 3.35 and 0 kg DM), respectively for diets I, II, III and IV. Diets were balanced for energy (2.53 Mcal ME/kg DM), aNDFom (34.8% DM), and CP (14.9% DM). Forage aNDFom (fNDF) was 22.5, 20.9, 19.5, and 18.0% and dietary starch was 22.6, 23.2, 23.8, and 25.1% DM, respectively. Cows were blocked by DIM (78±12), lactation n° (2.9±1.1) and ECM (46.8±3.6kg); they were housed in a tie stall and milked 3 times a day. Data were analysed using a mixed model. DMI among treatments I, II, III, and IV was 30.4, 30.4, 30.7, and 30.4 (p=.99); ECM was 43.2b, 43.7ab, 44.6a, and 44.2ab kg (p=.03). The sorting index of particles ≥6.7mm was 2.5b, 3.5ab, 3.1ab, and 4.7a (p=.04). Fecal production in 24h was 9.6a, 9.2ab, 9.4ab, and 8.9b kg of DM (p<.01), and total-tract digestibility of nutrients was only different for pdNDF240 (63.1a, 62.3a, 58.8b, and 58.1b % of pdNDF240; p<.01). Average reticular pH was 5.74 (p<.01). Average rumination time (RT) was 539min (p=.89); RT per kg of forage was 35.8a, 37.9bc, 42.6b, and 48.6a min/kg (p<.01), evidencing the lower fragility of WS compared to WH. The uNDF240 intake of diets was 3.7 kg (p=0.92). Results show that forage uNDF240 content and fiber digestibility rates are both important factors to be considered to manage feed intake and maintain proper rumen function of high-producing cows.

Effects of dietary supplementation of oregano essential oil on performance, rumen microbiota and morphometrics of feedlot cattle

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In ruminants, knowledge and ability to intervene in the dynamics of rumen microbiota is pivotal for feed efficiency. A proposed sustainable solution is the use of Oregano essential oil (OEO). Hence, our objective was to assess the impact of dietary supplementation of OEO on feedlot cattle performance, rumen microbiota and histomorphometry. Sixty-eight Limousine bulls were randomly assigned to 2 groups (n=34) designated as control (C) and test (T). Group C was fed a typical commercial diet whereas T was fed the same diet supplemented with OEO at 50mg/kg DM, for 90 days; feed intake was recorded weakly while body weight monthly. At the end of this period all bulls were slaughtered. Rumen fluid was collected for 16S rRNA gene sequencing. Histomorphological measurements of rumen tissue samples from each group were done using a computer-aided light microscope image analysis. Data were analyzed using SPSS software; significance level was set at P<0.05. Final body weight and average daily gain were similar between C and T. Bulls fed with OEO supplement had lower feed intake (P<0.05). Significant taxonomic differences were found within and between the two groups. Group T had a higher number of Succinivibrionaceae bacteria (P<0.05), that are linked to improved feed efficiency and higher expression of glutathione peroxidase (GPx) metabolic pathway (P<0.05). Rumen morphometric indexes differed between groups. Overall, the results showed significant effects on rumen microbiota and tissue. This research has been co-financed by the European Regional Development Fund of the European Union and Greek National Funds through the Operational Program Central Macedonia 2014-2020 (KMP6-0280294; BellasQualityMeat).

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Poster 14

Rumen fermentation and predicted methane emissions from dairy cows fed *Ascophyllum nodosum*

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The effect of feeding seaweed (*Ascophyllum nodosum*) to dairy cows on predicted methane (CH₄) production and rumen fermentation was assessed. 30 dairy cows were split into two groups (15 cows each) balanced for bodyweight, parity, and milk yield. Following an adaptation period of 4 weeks (wk) on a control diet (CON; total mixed ration, 48:52 forage:concentrate), each group continuously received for 9 wk: (i) CON diet, (ii) diet replacing 330 g/d of dried corn meal with 330 g/d of dried *A. nodosum* (SWD). Cows' dry matter intake (DMI) and milk yield was recorded daily. Milk and rumen fluid was collected in wk 3, 5 and 8. Volatile fatty acid (VFA) concentrations in rumen fluid were measured using gas chromatography and CH₄ production was predicted using published equations based on VFA. Data were analysed by linear mixed effects models with diet, wk, and their interaction as fixed factors; cow (nested within diet) as random factor. Acetic (P<0.05), butyric (P<0.001) and caproic (P<0.001) acids (mM) were higher (P<0.05) in SWD (74.6, 14.5, 0.7) than in CON (68.2, 11.9, 0.4). Predicted CH₄ production (g/d), yield (g/kg DMI) and intensity (g CH₄/kg energy-corrected milk yield) were higher (P<0.05) in SWD (517.1, 20.2, 16.4) than in CON (426.8, 18.5, 14.5). Replacing corn meal with *A. nodosum* increased predicted CH₄ emissions in dairy cows.

Effects of different feed processing forms on in vitro rumen fermentation characteristics

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The present study was conducted to evaluate the effects of processing corn on in vitro rumen fermentation with three different forms. The three treatments such as a basal diet with the ground, flaked, and pellet forms were arranged with in vitro rumen incubation technique, and its incubation times were 0, 2, 4, 6, and 12 h, respectively. Volatile fatty acid (VFA), ruminal pH, total gas production, ammonia nitrogen (NH₃-N), methane emission, and dry matter digestibility were analyzed to identify the rumen fermentation parameters. One-way ANOVA was used with the statistical analysis system R software in this experiment. Total VFA production was significantly increased ($p < 0.05$) by 7.1% in both ground and pellet treatments compared with flaked treatment at 4 and 12 h incubation. Acetate production was increased by 3.8% ($p < 0.05$) in both ground and pellet treatments in comparison to the flaked treatment during the whole incubation. Also, propionate production was higher at 6 h incubation in order for grind < flake < pellet, respectively ($p < 0.05$). All pH values were within the optimal range (5.8 – 7.2) for rumen normal microbial fermentation. Except for 4 h, the concentration of NH₃-N was increased in the ground treatment compared with other treatments during the whole incubation ($p < 0.05$). Similar to NH₃-N results, total gas production was significantly increased in ground treatment compared with other treatments at 6 h ($p < 0.05$). While, methane production was undesirably increased in ground treatment compared with other treatments during the whole incubation. The pellet form of corn is more degraded compared with both ground and flaked forms of corn ($p < 0.05$). Therefore, the present results suggest that the pellet form of corn is recommendable because no significant negative effects were observed on rumen microbial fermentation, and it could be expected to be available with rumen undegraded protein and acetate utilization.

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Poster 16

Effects of combination of garlic powder/attapulgit hybrid mixture and citrus by-product on performance and enteric methane production in Hanwoo steers

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In vivo evaluation of methane (CH₄) reducing feed additives is critical for practical implementation of effective enteric CH₄ reduction strategies in Korea. An in vivo trial was carried out by randomly allocating six Hanwoo steers (306 ± 17 kg) to three groups of two. The steers were fed commercial concentrate and Timothy hay (50:50) either supplemented with garlic powder/ attapulgit hybrid mixture: citrus by-product (50:50; GAC) at 2.0% DMI, or an un-supplemented diet (CON) in a triplicated 2 × 2 crossover design (two successive 25-day periods). CH₄ production was measured on day 21 for 3 days in each period using respiratory chambers. Rumen fluid and blood samples were collected on the last day of each period. Animals fed GAC exhibited 15.9% and 12.1% decrease in CH₄ production (122.8 vs. 103.3 g/d; $p < 0.05$) and yield (18.7 vs. 15.1 g/kg DMI; $p < 0.05$), respectively, when compared to CON. However, a decrease in DMI (7.8 vs. 7.1 kg/d; $p < 0.05$) followed by drastic decrease in average daily gain (0.70 vs. 0.35 kg; $p < 0.05$) was observed in GAC, perhaps due to the decreased palatability caused by top-dressing GAC on to the diet. Ruminal pH, ammonia and total volatile fatty acid concentration did not differ between the treatments ($p > 0.05$), but GAC supplemented diets slightly increased ($p = 0.069$) the acetate:propionate ratio. Analysis of blood serum metabolites revealed elevated ($p < 0.1$) bilirubin and alkaline phosphatase levels in GAC fed animals. Overall, although GAC can decrease CH₄ emissions, at the tested dosage, it may cause adverse effects on productivity and health status of the animals. Future studies are needed to optimize the effective dosage level and method of supplementing GAC to the animals.

Short-chain fatty acids transporters expression in ruminal epithelium of growing rams depends on the source of butyrate used in a diet

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High intake of concentrate and resulting high short-chain fatty acids (SCFA) production in the rumen may exceed the ruminal epithelium ability to absorb them. In order to enhance ruminal epithelium function, butyrate supplementation in the diet can be used. The aim of the study was to compare two sources of dietary butyrate: commonly used sodium butyrate (SB) and tributyrin (TB), as an alternative to SB, when those were supplemented in a high-concentrate diet of growing rams. Thirty-two rams (30.6 ± 2.5 kg; 11-14 months of age) were allocated into one of four treatments and fed ad libitum diet with: 1) low inclusion of concentrate (22.5% of diet dry matter (DM); L); 2) high inclusion of concentrate (60% of DM; H); 3) H with SB (3.2% of DM; H+SB); and 4) H with TB (2.93% of DM; H+TB). After three weeks, the rams were slaughtered and SCFA transporters mRNA expression in ruminal epithelium was analyzed. Targeted genes included: monocarboxylate transporter 1 (MCT1), putative anion transporter-1 (PAT1) and downregulated in adenoma (DRA). The pre-planned contrasts were used for data analysis: L vs. H, H+SB and H+TB; H vs. H+SB and H vs. H+TB. In atrium ruminis, DRA expression was higher for L compared to H treatments (P=0.03). In ventral rumen, MCT1 expression was lower for L compared to H treatments (P=0.02). When it comes to butyrate sources, H+SB compared to H treatment tended to increase PAT1 (P=0.09) and decrease DRA (P=0.06) expression, while TB had no impact on investigated transporters mRNA expression (P≥0.14). Unlike SB supplementation, the TB supplementation does not affect the SCFA transporters expression in the ruminal epithelium.

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Poster 18

Relationship between the natural ¹⁵N enrichment of plasma and feed conversion efficiency in fattening young bulls

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The natural ¹⁵N enrichment of animal proteins over the consumed diet ($\Delta^{15}\text{N}$) has been related to feed conversion efficiency (FCE) in beef cattle and therefore can therefore be used as a proxy for selection and classification of animals. The relationship between $\Delta^{15}\text{N}$ measured in the whole plasma and FCE has been studied in beef fattening young bulls under different feeding regimes (2 concentrates x 2 forages). The $\Delta^{15}\text{N}$ values from plasma samples of 52 male calves were obtained at 117 d (M1) and at 201 d (M2) of age. Daily body weight and concentrate intake was obtained from automatic scales and feeders, respectively. Individual forage intake and faecal production was estimated with double marker from faecal samples, while excreted N was estimated from spot urinary creatinine and N contents of urine and faeces. The FCE on M2 was calculated as average daily gain (from 173 to 229 days of age) divided by total DM intake (FCE_DM) or by retained N (FCE_N). The $\Delta^{15}\text{N}$ values in plasma samples at M1 were poorly and negatively correlated with FCE_DM (-0.32, p<0.05) and FCE_N (-0.22, P<0.05) whereas the $\Delta^{15}\text{N}$ values of the samples at M2 were moderately and negatively correlated with FCE_DM (-0.61, p<0.05) and FCE_N (-0.61, P<0.05). Animals were classified into three groups according to its FCE on M2 (HIGH_FCE, MED_FCE, and LOW_FCE). Animals belonging to HIGH_FCE had lower $\Delta^{15}\text{N}$ in M1 than LOW_FCE animals for both FCE_DM (4.30‰ vs 4.52‰, P<0.05) and FCE_N (4.34‰ vs 4.50‰, P<0.05) traits. In the case of $\Delta^{15}\text{N}$ in M2 the difference was only observed for FCE_DM. In conclusion, our finding confirms that the natural ¹⁵N enrichment of plasma over the diet is a predictor of feed efficiency but the relationship depends on the specific used trait.

Effects of feeding rumen-protected lysine on milk performance in lactating Holstein dairy cows

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Lysine (Lys) has been identified as a limiting amino acid (AA) for milk and milk protein production in lactating dairy cows. A 30-d pen study was conducted in a commercial Parmigiano-Reggiano to evaluate the effect of supplementing a rumen-protected lysine product (Relys®50; Vetagro S.p.A.; Reggio Emilia, Italy) on lactation performance in high-yielding dairy cows. One hundred ninety-five multiparous Holstein cows (3 ± 1.1 lactations [mean \pm SD], and 150 ± 85 days in milk [DIM]) were randomly assigned to one of two groups: Control ($n = 95$; CON; basal diet) and RPL ($n = 100$; CON + 45 g/d of Relys®50 [50% HCl-Lysine]). Basal diet was supplemented with rumen-protected methionine (30 g/d of Timet®; 55% DL-methionine [Met]; Vetagro S.p.A) to provide 1.14 g of Met/Mcal metabolizable energy (ME). Respectively, CON and RPL diets were formulated at 2.86 and 3.04 g of Lys/Mcal ME, and 2.5:1 and 2.7:1 Lys:Met ratio. Cows were housed in free-stall pens and milked twice a day. Milk yield was recorded daily, and milk components were analyzed once a week. Data was divided by stage of lactation according to DIM (early [0-35], peak [36-90], mid [91-150], and late [151-305]) and analyzed under a mixed model with the random effect of cow and the fixed effects of lactation, season and DIM, treatment and their interaction. Supplementation of RPL increased milk yield in mid ($P=0.12$) and late ($P=0.03$) lactation compared to CON. In early lactation cows, milk protein and casein tended to increase in RPL compared to control cows ($P<0.15$). Overall, milk casein yield increased 40 g in RPL cows compared to CON ($P<0.15$). Milk urea nitrogen was significantly reduced in peak and mid lactating cows fed RPL relative to CON ($P<0.05$). Differential somatic cell count was lower in RPL cows across all lactation compared to CON ($P<0.15$). We conclude that feeding Relys®50 is a valid strategy for AA balancing and positively affects milk performance and nitrogen utilization in lactating dairy cows.

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Poster 20

Evaluation of plasma parameters in fattening beef cattle fed hexane vs 2-methyloxolane defatted soybean meal

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Chemical extraction of fat to produce soybean meal (SBM) commonly uses solvents like extraction-grade hexane (Hex), which poses potential toxicity risks. Concerns over health risks have prompted the exploration of alternatives such as 2-methyloxolane (2-meOx), a bio-based solvent derived from agricultural by-products. This study aimed to evaluate plasma parameters related to liver and kidney function in fattening beef cattle fed over the long-term diets containing SBM extracted using 2-meOx compared to hexane-extracted SBM, with or without supplementation of rumen-protected methionine (RPM). Thirty-six young Charolais bulls (248 ± 21.7 days) were studied during three consecutive 70-day feed efficiency tests. Treatments were determined by a factorial design crossing two types of SBM (Hex vs 2-meOx) with 2 levels of RPM supplementation (with vs without). Blood samples were collected on days 44, 100, and 156. Blood plasma underwent spectrophotometric analysis for glucose, β -hydroxybutyrate, NEFA, urea, creatinine, total plasma proteins, albumin, cholesterol, triglycerides, ALT, AST, GGT, and ALP. The RPM supplementation did not significantly affect plasma parameters, except for a higher urea concentration observed when Hex SBM was supplemented with RPM ($P = 0.02$). The only two plasma parameters differing between Hex and 2-MeOx diets were the ALP ($P = 0.02$) and GGT ($P = 0.07$) enzymes, both exhibiting higher values in the Hex group at all three sampling points. Although these results should be complemented by additional studies, they may suggest potential chronic and subclinical liver damage in beef cattle fed SBM extracted with Hex compared to 2-meOx.

Modulatory effects of HMTBa and sea buckthorn oil on milk fat depression in dairy sheep

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Ewes are not prone to milk fat depression (MFD) due to dietary starch and plant oils, as opposed to cows, but fish oil induces MFD in both species. The origin of fish oil-induced MFD is unclear, as it cannot be explained only by increases in t10 fatty acids (FA). Thus, it has also been attributed to FA from fish oil having direct antilipogenic effect, in particular c9-16:1. On this basis, we conducted a trial to examine the ability of 2-hydroxy-4-methylthiobutanoate (HMTBa; an additive that mitigates t10 production) and sea buckthorn oil (SBO; rich in c9-16:1) to modulate MFD in ewes. The trial followed a replicated 3×3 Latin square design (n=12 ewes) with 3 periods of 25 days and 3 diets: a ration including 1.5% DM fish oil to induce MFD (control), and the same diet supplemented with 0.1% DM HMTBa or with 1.5% DM SBO. Milk yield and composition (including a detailed FA profile) were analyzed on the last 3 days of each period. Contrary to expectations, HMTBa did not affect (P>0.10) the proportions of t10-18:1 and t10c12-18:2 in milk, or milk fat content and yield, which contrasts with positive responses to ≤0.1% HMTBa in cows suffering from MFD due to high-starch diets and plant oils. We speculate that ruminal bacteria involved in the t10 shift induced by fish oil may be unresponsive to HMTBa or a higher dosage is needed. The apparent transfer of c9-16:1 from SBO to milk averaged 16% (≈122% increase in milk; P<0.01), but this FA failed to modify milk fat content, thus dismissing an antilipogenic action in ewes. Overall, HMTBa and SBO failed to modulate MFD in sheep. Acknowledgment: PID2020-113441RB-I00 and PRE2021-098235 (MCIN/AEI/Spain).

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Poster 22

Impact of Tall Fescue defoliation intensity on energy expenditure of mid-lactation dairy cows

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To quantify the effect of grazing intensity on energy partitioning of mid-late lactation of fall-calving dairy cows (224 ± 7 days in milk), multiparous Holstein cows (n = 36; 16.7 ± 0.9 kg of milk and 633 ± 69 kg of body weight; BW) were used. Cows grazed a Fescue pasture (16 h of access) starting when three new extended leaves developed until reaching 9 (TC), 12 (TM) or 15 (TL) cm of post-grazing sward height. Cows were milked twice a day and milk samples were collected for composition analysis (fat, protein, and lactose); BW and body condition score were recorded. Heat production (HP) throughout the day was measured using the Heart Rate (HR)–O₂ pulse technique (Brosh, 2007). Data were analyzed with a mixed model and means were declared different when P ≤ 0.05. Retained energy (RE) in milk was greater for TL than for TM and TC cows and for TM than TC cows (435 vs. 352 vs. 287 ± 28 kJ/PV_{0.75} per day, respectively), while metabolizable energy intake (MEI) tended (P < 0.10) to be higher for TL and TM than TC (1435 and 1460 vs. 1226 ± 124 kJ/PV_{0.75} per day for TL, TM and TC, respectively). Heat production did not differ among grazing intensities and averaged 984 ± 335 kJ/PV_{0.75} per day. The gross energy efficiency (RE-milk/MEI) tended (P = 0.06) to be greater for TL than TM and TC cows (0.30 vs. 0.24 and 0.25, respectively). Daily HP along the five days of grazing in the same paddock showed that there was no effect of the day of grazing or its interaction with treatment. Results suggest that although daily HP did not change between the different treatments and as herbage height decreased as cows grazed in the same paddock, energy expenditure for grazing was probably less for TL which explained its greater energy efficiency.

The effect of energy and nitrogen source and their interaction on rumen fermentation and methane formation in a RUSITEC system

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An in vitro experiment with the rumen simulation technique (RUSITEC; three 10-day runs) was used to assess the effect of energy (E) and nitrogen (N) sources on rumen fermentation and CH₄ formation. Purified carbohydrate and N sources were used with pectin (Fibre) or corn starch (Starch) as the main E source, combined with ammonium sulphate (AS), casein (CAS), amino acid (AA) mixture 1 (AA1) or AA2. A total of 20.6 g of feed dry matter (DM) was fed per day; inoculum and gas samples were collected during the last 5 d. Data were analysed with a mixed model in R, with effect of E and N source, and E × N as fixed and run as a random effect. Total volatile fatty acids (VFA) and total gas production were not affected by treatment, and pH was lower ($P < 0.01$) for AS and AA compared with CAS, and lower ($P < 0.01$) for Starch vs. Fibre. Both E and N sources affected ($P < 0.05$) NH₃ concentration, being greater for Fibre than Starch (15.3 vs. 14.8 mM), and of N sources greatest for AS (28 mM) and lowest for CAS (1.9 mM). Overall, the purified feeds favoured butyrate (52.0 ± 6.59 mol-%) over propionate (2.21 ± 0.398 mol-%) production. There was an E × N interaction ($P < 0.01$) for acetate and butyrate mol-%, with acetate being greatest and butyrate lowest for Starch with AS. Lastly, CAS had greater CH₄ yield (mL/g DM; $P < 0.01$), regardless of E source, compared with other N sources, being 81%, 86%, and 74% lower for AS, AA1 and AA2, respectively. Further analysis of E and N utilisation for microbial protein synthesis and microbial populations will confirm the observed effects on rumen fermentation.

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Poster 24

Evaluation and development of analytical procedures to assess buffering capacity of ruminant feed buffers
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The inclusion of rumen buffers in ruminant feed prevents rumen acidosis, avoiding the negative production and health consequences of low rumen pH, consequently improving feed efficiency. Benchmarking the efficacy of rumen buffer materials is of significant interest to feed mills and producers. The aim of this study was to evaluate, develop and optimise laboratory protocols to consistently and robustly evaluate rumen buffering materials to provide confidence in their in vivo efficacy. 3 different methods were evaluated for determining buffering potential of materials: a) 2 and 8 hour pH STAT, b) 8 hour fixed HCl acid load addition and c) 3 hour acidotic diet simulation using acetic acid. The uncertainty of measurement (UoM) of the pH STAT titration methodology was determined at pH 5.5 and 6.0 and was found to range from 0.51-4.39 and 0.93-2.55 ml of HCl, respectively, for 5 different carbonate-based buffer materials. The fixed HCl acid load methodology was evaluated to determine the method variables that have a significant effect on the buffer potential to prevent subacute rumen acidosis, reported as area under the curve (AUC, mmol H⁺. s). Buffer material, threshold pH, test duration and interactions between all 3 variables were significant ($p < 0.001$) in predicting the performance of the buffer materials. The acidotic diet simulation was found to provide a different ranking of materials to the 8 hour fixed HCl acid load methodology. The results highlight the importance of method selection and test parameters for accurately evaluating the potential efficacy of rumen buffer materials.

An in vitro alternative to the in sacco method to determine ruminal starch degradation

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Starch is a pivotal constituent of many ruminant feeds, serving as a key energy source. Determining its degradation in the rumen is essential, as it enables to calculate the risk of acidosis and indirectly determines the partitioning of energy digestion between the rumen and the intestine, thereby influencing nutrients intake. The part of degradable starch in the rumen used to be determined on animals with the in sacco method. However, societal demand on animal welfare led to the development of in vitro methods to determine ruminal degradation of feeds, recognizing as well the significant cost and time savings offered by laboratory methods. An enzymatic method was developed to estimate starch ruminal degradation and tested on 17 common concentrated feed used in ruminant feeding (cereals, by-products, feed mixtures and legume seeds). Starch content of feed varied from 197 to 682 g/kg DM. The method consists of incubating dried and ground samples, at 60°C in buffer pH6 for one hour with a *Bacillus* sp. alpha-amylase (10069, Sigma-Aldrich, France). Enzymatic starch degradation was calculated as the ratio between the quantities of starch degraded (recovered from the residue after incubation) and the initial starch content of feed. This enzymatic in vitro starch degradation was compared to the in sacco ruminal degradation (assuming passage rate of 6%/h) of starch (ED6_St). ED6_St varied from 64% to 97% and enzymatic degradation varied from 39 to 96%. The coefficient of determination between the enzymatic method and the ED6_St was 69.9%, and the residual standard deviation was 6.3%. These first results are promising to obtain an in vitro method to assess ruminal starch degradation. However, further tests are required to complete and validate the method, including the incorporation of a broader array of feed types, such as forages.

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Poster 26

Meta-analysis explores factors affecting milk component responses to metabolizable methionine fortified evaluated using NRC2001

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Objective is to establish response equations on milk protein and fat contents and yields (MPC, MFC, MPY, and MFY) to rumen-protected methionine (RPM) supplementation as Smartamine M or MetaSmart and identify influencing factors. Meta-analysis applied to peer-reviewed data (26 publications). Lactating cows consumed 22.7±3.9kgDM, 160.2±15.8g CP/kgDM, 1.59±0.1 McalNE/kgDM and 2.14±0.4 mMet%MP. Robust within-study models predicted MPC, MPY, MFC and MFY with 0.89, 0.94, 0.89 and 0.97 R²adj with 0.05%, 45.0g/d, 0.14% and 59.0g/d, respectively, to RPM based on metabolizable methionine (mMet) as % metabolizable protein (%mMet). MPC and MPY models suggested a 0.6% mMet increase resulted in a 0.14% and 54g/d increase. Within-study slopes of MPC and MPY were positively impacted by mLys/mMet, %mHis (P<0.01), NEL, EE, %mArg (P<0.05) and %mVal (P<0.1) and negatively impacted by %mLeu (P<0.1). Within-study slopes of MPC were positively impacted by bacterial MP (P<0.05). For the same %mMet content, MFC increased with RUP, %mArg, %mIle, %mThr and decreased with %mLeu (P<0.05) where MFY increased with DMI, EE, NEL, Bacterial MP, MP, RDP, RUP, %mHis, and mLys/mMet and decreased with %mLeu (P<0.05). MFC and MFY models indicated an increase of 0.14% and 66g/d, by increasing 0.6% mMet. Within-study slopes of MFY were positively impacted by EE, %mHis, %mVal and mLys/mMet (P<0.05) and negatively impacted by %mIle (P<0.05). For the same %mMet, MFY increased in early lactation, DMI, EE, NEL, Bacterial MP, %mHis, mLys/mMet (P<0.05) and decreased with %mLeu (P<0.05). MFC decreased with %mLeu (P<0.05). Predicting lactating dairy cows' responses to RPM and understanding influencing factors is feasible, enhancing comprehension and aiding dietary formulation improvements.

Meta-analysis explores factors affecting milk component responses to metabolizable methionine fortified evaluated using CNCPS

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Objective is to establish response equations on milk protein and fat contents and yields (MPC, MFC, MPY, MFY) to rumen-protected methionine (RPM) supplementation as Smartamine M or MetaSmart and identify influencing factors. Meta-analysis applied to peer-reviewed data (26 publications). Lactating cows consumed 22.5 ± 3.9 kg DM, 161.5 ± 18.6 g CP/kg DM, 2.59 ± 0.1 Mcal ME/kg DM, and 1.04 ± 0.2 g mMet/Mcal ME. Robust within-study models predicted MPC, MPY, MFC and MFY with 0.89, 0.94, 0.88 and 0.97 R^2_{adj} and 0.05%, 45.0g/d, 0.15% and 57.0g/d RMSE, to RPM based on metabolizable methionine as g/Mcal metabolizable energy (mMet, g/Mcal ME). MPC and MPY models showed a 0.29g/Mcal ME mMet increase resulted in a 0.14% and 51g/d increase. Within-study slopes were positively impacted by EE, mHis (%MP), and mLys/mMet ($P < 0.05$) for MPC and MPY and by ME, mLys (%MP), mVal (%MP) for MPY ($P < 0.05$). For the same g/Mcal ME mMet, MPC decreased with DMI, Starch, ME, mLeu (g/Mcal ME), and increased ($P < 0.05$) with MP, RUP, mArg, mLys, mIleu, and mThr (as %MP), where MFY increased in early lactation, DMI, ME, MP, EE ($P < 0.05$), mHis (g/Mcal ME), mLys/mMet ($P < 0.1$), and decreased with mIle (g/Mcal ME, $P < 0.1$). MFC and MFY showed a 0.16% and 66g/d increase with a 0.29g/Mcal ME mMet increase. Within-study slopes were positively impacted by ME for MFC ($P < 0.1$) and by lactation stage and mLys/mMet for MFY ($P < 0.01$). For the same g/Mcal ME mMet, MFC increased in early lactation ($P < 0.05$) and decreased by ME, RUP, mLy (%MP), mHis, mIle, mLeu, mPhe, mVal (g/Mcal ME, $P < 0.05$), where MFY increased in early lactation and by ME ($P < 0.05$) and decreased by mIle (g/Mcal ME, $P < 0.01$). Predicting lactating dairy cows' responses to RPM and understanding influencing factors is feasible, enhancing comprehension and aiding dietary formulation improvements

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Poster 28

Energy requirements are affected by posture and activity in dairy cows

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Twenty-two multiparous Holstein cows (656 ± 59 kg BW; 92 ± 14 DIM; 35.6 ± 5.4 kg of milk/d) were used to quantify the impact of posture and activity on their energy requirements. Cows milked twice daily and grazed a *Lolium arundinaceum* pasture (0800 to 1400 h) and were supplemented with a TMR (55% of the predicted DMI) in a compost bedded pack barn (1600 to 0400 h). Heat production (HP) throughout the day was measured using the heart rate-O₂ pulse technique (Brosh, 2007), cow posture (lying down or standing) was recorded using a three-axis accelerometer attached to the medial side of the hind leg (Ledgerwood et al., 2010), and cow activity (eating, grazing, ruminating or idling) was recorded via halters containing a noseband pressure sensor and a three-axis accelerometer (RumiWatchSystem, ITIN+HOCH) for 5 days. The combination of lying down and idling resulted in the lowest HP (866 ± 72 kJ/kg_{0.75}/d) and HP increased by 66 ± 3 kJ/kg_{0.75}/d when cows were standing compared to lying down. On the other hand, HP increased 15 ± 2 kJ/kg_{0.75}/d when cows were ruminating compared to idling whereas grazing on pasture or eating at the feed bunk led to increments of 121 ± 5 or 163 ± 4 kJ/kg_{0.75}/d compared to idling, respectively. Based on this information, ruminating would not represent an important energy cost for cows, but feed consumption (grazing and eating) incurred the greater energy cost for cows. The HP of eating at the feed bunk was 35% greater than grazing on pasture, which is related to a greater feed intake rate and, consequently, a higher heat increment. This study demonstrates that considering activity and posture throughout the day can enhance the estimation of the energy requirements of dairy cows.

Metabolic status of feed restricted grazing dairy cows during early and mid-lactation

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To evaluate the metabolic adaptation of grazing cows to a 5-day feed restriction period during early (31±14 days in milk; DIM) and mid-lactation (146±17 DIM), multiparous Holstein cows randomly assigned to two treatments: ad libitum (ADLIB; n=12) or restriction (RESTR: 50% of ADLIB offered dry matter, DM; n=12). In each stage of lactation, cows were evaluated in three periods: an adaptation (fed ADLIB; 8 days), a treatment (5 days, TREAT) and a flushing (fed ADLIB; 8 days; FLU) period. Pasture DMI was 17.0±2.4 vs. 9.4±1.1 kgDM/d and 9.0±4.4 vs. 11.5±1.4 kgDM/d for ADLIB vs. RESTR cows during early and mid-lactation, respectively. Supplementation with an energy concentrate at the milking parlor represented 20% of total DMI. In addition, during mid-lactation ADLIB cows were offered 9.5±1.7 kgDM/d of ryegrass haylage due to low pasture availability. Blood samples were collected twice during every period. Data were analysed with a mixed model and means differed when $p < 0.05$. Both, energy corrected milk (ECM) yield and body condition score (BCS) decreased for RESTR than ADLIB cows during TREAT in early and mid-lactation, being ECM yield, but not BCS, recovered during FLU. Non-esterified fatty acid and beta-hydroxybutyrate concentrations increased during TREAT for RESTR than ADLIB cows only during early lactation. However, plasma glucose was reduced while albumin and aspartate aminotransferase ($p=0.06$) were increased for RESTR than ADLIB during TREAT and FLU in early and mid-lactation. Cholesterol, insulin to glucagon ratio, urea and creatinine concentrations were not affected by feed restriction. Feed restriction during early lactation resulted in greater lipid mobilization and although feed restriction during mid-lactation decreased ECM overall metabolic status was more favorable during this stage.

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Poster 30

Evaluating the relationship between undigestible neutral and acid detergent fiber and implications for cell wall components in forages

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Fiber digestibility, notably knowing undigestible neutral detergent and acid detergent fiber (uNDF, uADF), is vital for understanding rumen dynamics. The relationship between uNDF and uADF is not well explored and might provide insight into factors related to end-product formation, cross-linking, and methane production. Our aim was to determine how much uNDF residue was explained by uADF residue. A total of 14 forage feeds were analyzed, including 3 alfalfa hays, 4 corn silages, 4 grass silages, and 3 perennial pasture ryegrasses. Samples were analyzed for NDF and ADF and their respective residues measured after in vitro fermentation for 240 h. Other nutrient composition in feeds were determined using near infrared spectroscopy. Data were analyzed using the regression procedure of SAS. We observed a strong relationship between uNDF and uADF (adjusted [adj] $R^2 = 0.99$). On average, uADF represented 78% of the uNDF among samples. Forage type might influence this relationship; uADF represented 94.5% of uNDF variation in alfalfa hay and 66.6% in perennial ryegrass. Total non-digested hemicellulose content appeared to be a better predictor of both uNDF and uADF (adj $R^2 = 0.70$ and 0.75) when compared to cellulose (adj $R^2 = 0.30$ and 0.31). Variability associated with cellulose digestion among feeds could account for this reduced predictability (e.g., 86% for perennial ryegrass vs. 42.2% for alfalfa hay), but also the level of cross-linkage among forages. Overall, the observed relationship between uADF and uNDF deserves further investigation as it relates to both animal productivity and enteric methane production.

Post-grazing sward height affects resting time on pasture

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To evaluate the effect of the post-grazing sward height on the resting time at pasture, thirty-two multiparous Holstein cows (679 ± 59 kg BW, 186 ± 14 days in milk) were assigned to 2 treatments (6 vs. 14 cm post-grazing sward height; T6 or T14, respectively; 4 cows/treatment/paddock). Cows grazed (from 0800 to 1400 h) a *Lolium arundinaceum* pasture with a pre-grazing sward height of 20 cm and within treatment grazed in the same paddock (5 days) until reaching 6 or 14 cm. Cows were supplemented with a TMR (55% of the predicted dry matter intake) in a compost bedded pack barn (from 1600 to 0400 h). Resting time was recorded using a three-axis accelerometer attached to the medial side of the hind leg. Data were analyzed in a mixed model with treatment, day of occupation and its interactions as fixed effects and paddock as a random effect. Milk yield was greater while resting time on pasture was lower for T14 than T6 cows (25.8 vs. 23.9 ± 0.8 kg/d and 111 vs. 95 ± 8 min/d, respectively). These results suggest that T14 cows spent less time grazing than T6 cows and therefore the higher post-grazing sward height would have allowed cows to achieve a higher forage intake rate and with probably also a higher digestibility which led to a greater digestible nutrient intake and thus, milk production. Independently of treatment, milk yield was greater on days 3 and 5 than day 1 of paddock occupation, which would probably be associated to the lowest nutrient intake on the last day of occupation on the previous paddock. However, resting time was not affected by the day of occupation indicating that as pasture height decrease, high supplemented cows would not increase their grazing time.

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Poster 32

Effects of anthocyanin-rich Napier grass silage in growing goats

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The purpose of the study was to find out how goats that are growing respond to silage made from anthocyanin-rich Napier grass. Three groups of eighteen goats were randomly assigned: (1) Napier grass silage (negative control: T1); (2) anthocyanin-rich Napier grass silage without additive (positive control: T2); and (3) anthocyanin-rich Napier grass silage with 4% molasses and 0.03% FeSO₄ (treatment: T3). The findings showed that the goats fed T3 had a significant ($P < 0.05$) increase in their intake of crude protein and other nutrients. The digestibility, ammonia nitrogen concentration at 4 hours, nitrogen balance, blood urea nitrogen, average daily increase, body weight gain, total volatile fatty acid, and body weight were all significantly greater ($P < 0.05$) in the T3 diet. Nevertheless, no appreciable variations were noted for pH value and population of microorganisms in every group. According to the study's findings, utilizing anthocyanin-rich Napier enhanced non-enzymatic activity and decreased lipid oxidation in the plasma of developing goats. Napier grass silage, which is high in anthocyanins, can be fed to animals to help them reduce the negative effects of oxidative stress from the surroundings.

Comparison of dietary digestibility of dairy ewes and goats during early lactation

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Feed intake and nutrient digestibility of Sarda ewes and Saanen goats were studied simultaneously to assess possible species differences in early lactation. Ten animals of each species (40 ± 3 days in milk and 47.4 ± 5.3 kg of body weight (BW) for sheep, 37 ± 4 days in milk and 57.8 ± 7.8 kg of BW for goats) were put into individual metabolic cages and fed ad libitum diet containing 20.4% starch, 35.4% NDF, 16.2% CP, on a DM basis. During the trial, milk, ort, rumen fluid, feces, and urine samples were collected. Data were analyzed by the PROC GLM procedure of SAS to test the differences between species. Dry matter intake (DMI) and milk production were higher in goats than in ewes (3.09 vs. 2.35 ± 0.36 kg/d; $P=0.051$; 4.31 vs. 2.23 ± 0.43 kg/d; $P=0.0001$), whereas DMI expressed in % of BW did not differ between the two species. Feed energy efficiency (ME energy in the milk/total ME intake) was significantly higher in goats (0.65 vs. 0.53 ± 0.08). Dry matter (DM) and CP apparent digestibility, starch and NDF digestibility, and total digestible nutrients (TDN) did not differ between the two species, while NFC apparent digestibility was higher in goats than in ewes (98.46 vs. 91.74 ± 0.33 %; $P<0.001$). Goats had higher rumen pH (5.86 vs. 5.70 ± 0.07), ammonia concentration (23.38 vs. 11.89 ± 3.46 md/dl) and microbial protein flow (130.64 vs. 90.72 ± 14.62 g/d) than ewes. In conclusion, sheep and goats fed in the same exact animal and dietary conditions did not differ in their levels of intake and digestibility, except for NFC digestibility. Goats, however, had higher rumen pH, confirming their ability to manage starch rich diets, and transformed feed energy in milk energy more efficiently than ewes (Funded by LR 7/10/2007, n. 7, ann. 2012).

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Poster 34

The supplementation with Omega 3 fatty acids enhances the reproductive performance in dairy cattle

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The aim of this study was to estimate the impact of the supplementation with Omega 3 fatty acids source (Hi-flax) on the reproductive performance in dairy cattle. Data was collected in a commercial farm located in Department Ille et Vilaine (France) from March 2022 to January 2023. Cows were randomly assigned to two treatments: T1 (n=15), a basal diet, and T2 (n=16), T1 with a daily supplementation of 250 g/cow of rumen protected flaxseed oil (Hi-Flax) in the first 45 days post-calving. Analysed parameters were the conception per artificial insemination (CR: 0, non-pregnancy and 1, pregnancy), the number of insemination (N.IA) until pregnancy and the days from calving to successful insemination (OD, days). Data were analysed using lmer R package from R software (R Core Team, 2021). For CR, generalized linear mixed model effects with repeated measures was performed assuming a Binomial distribution for the response variable, where the number of insemination and treatment were included as fixed effects and the cow as random effect. For the OD and N.IA, generalized linear models were performed (non-repeated measures) with number of insemination and treatment as fixed effects and for N. IA, only treatment was considered as fixed effect. The results showed a superiority in CR for T2 with a log ODDs ratio of 2.45 ($p=0.08$), and an increase from 0.373 to 0.596 (+60%) in the term of probability ($p=0.08$). Secondly, N.IA were lower for T2 in 0.5, from 2.21 to 1.70 ($p=0.06$). Finally, OD was lower in 32d (25%, $p=0.01$) for T2. Interpretation suggests an enhancement in cow fertility, probably associated to the upregulation of PG3 α production. This biological mechanism is implicated in bolstering corpus luteum and follicular development while concurrently diminishing the PG2 α signal. Consequently, such physiological enhancements involve cost savings due to reduced veterinary, labour, and management costs, alongside decreased non-productive and dry periods, ultimately culminating in an improvement in overall profitability for dairy farm. In summary, this study underscores the benefits of Hi-Flax supplementation, with a significant improvement in reproductive performance and, by extension, profitability of dairy farming.

Feeding dairy cows in the morning or in the evening during the summer season: effects on milk production, feeding behavior, and rumen activity

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Night feeding has been proposed as a solution to mitigate the impact of heat stress in dairy cows during summer. In this study, we assessed the effects of this practice on milk production and behavioral traits. During summer, 40 lactating Holstein cows were enrolled in a cross-over design with 14-d periods and were either fed in the morning (0800 h; MF) or in the evening (2000 h; EF). In-barn THI was continuously monitored and respiration rate was measured for every cow twice daily. Milk production, dry matter and water intakes, rumination and lying times, and rumen temperature were automatically recorded throughout the day, and feeding behavior was characterized. Data were analyzed with repeated measures mixed models (proc GLIMMIX of SAS). The average hourly THI was always beyond 68, a threshold commonly used to define heat stress. Daily milk yield was not affected by feeding time but EF produced more milk than MF in the afternoon. Daily DMI was unaltered, but MF had a greater daily eating time, lower meal frequency, and longer average meal length. Although MF tended to have a greater average meal size, EF ate more feed in the conditioned meal occurring immediately after feeding, with only minor differences in intake patterns during the rest of the day. Daily rumination and lying times were similar, but rumination was greater in MF around milking times. Moreover, EF cows were more active overnight, with decreased lying time and increased activity relative to MF. Respiration rate was higher in EF in the morning, consistent with the greater rumen temperature observed in those cows from 0500 to 1500 h. Overall, night feeding did not improve heat stress response but altered the daily patterns of several behavioral traits.

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Theatre 2

Effects of residual sward height on grazing behaviour and milk production of dairy cows

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The impact of two defoliation intensities on a *Lolium arundinaceum* pasture on dairy cows' grazing behaviour and milk production was studied in Uruguay. Two treatments, Traditional and Lax, maintained residual sward heights of 5-7 cm and 12-15 cm, respectively. Thirty-two autumn-calving multiparous Holstein cows were allocated to 1.6-ha paddocks in a randomized complete block design with four spatial replicates. The same cows were used throughout the study and were kept under rotational grazing (8:00 to 14:00 h). During two 5-day occupation periods, daily grazing time, first grazing meal length, and mean grazing meal length were recorded. Milk production was registered twice daily. Cows consumed 12.4 ± 0.62 kg/d of a partial mixed ration with a 69:31 concentrate: sorghum silage ratio. Data were analysed with a mixed model including treatment, moment of the occupation period (beginning, days 1-2; middle, day 3; and end, days 4-5) and their interactions as fixed effects and the 5-day occupation period as random effect. Pre- and post-grazing sward heights were 21.4 ± 2.8 cm and 7.7 ± 1.2 cm for Traditional, and 22.5 ± 0.6 cm and 16.0 ± 0.9 cm for Lax treatment. Daily grazing time was not affected by treatment but increased from beginning to end of occupation periods (290 vs 304 ± 7.6 min/cow, $P = 0.04$). In Traditional, both first grazing meal length (174 vs 139 ± 8.5 min/cow; $P = 0.04$) and meal length (119 vs 96 ± 6.9 min/cow; $P = 0.02$) were higher compared to Lax treatment. Milk production was 33.3 ± 1.27 and 37.4 ± 1.30 kg/cow/day ($P < 0.01$) for Traditional and Lax treatment, respectively, decreasing by 1.4 ± 0.34 kg/cow/day from beginning to end ($P < 0.01$) of the occupation period. No significant interactions were found. In conclusion, milk production was greater with Lax than Traditional treatment, associated with changes in ingestive behaviour.

Effect of feeding system on lying behavior of mid-lactation dairy cows

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The effect of feeding strategy and environmental control during supplementation was evaluated using spring calving multiparous Holstein cows during two years (40 and 36 cows, respectively). Cows within year were blocked by calving date body weight (653 ± 74 kg) and body condition score (3.0 ± 0.2) and assigned in a randomized block design to three feeding systems: a confined 100%-fed total mixed ration (CB-TMR) or two mixed systems in which cows grazed and received supplementation confined in a compost-bedded pack barn (CB-GRZ) or in an outdoor soil-bedded pen (OP-GRZ). Cows were milked twice a day (04:30 and 16:30 h) and milk yield was recorded. At 165 ± 13 days in milk (during summer), cow lying time was recorded using three-axis accelerometers attached to the medial side of the hind leg, for 5 days. Milk yield was 30% greater ($p < 0.01$) and lying bouts were 10% more frequent ($p = 0.06$) for CB-TMR than for CB-GRZ and OP-GRZ cows (38.7 vs. 26.0 ± 3.9 kg/d and 11.5 vs. 10.3 ± 0.5 bouts/d). However, lying time per bout did not differ due to treatment and averaged 52 ± 2 min/bout. Differences in lying behavior between CB-TMR and mixed systems were observed at night hours (sunset to sunrise) when cows in mixed systems grazed; both, CB-GRZ and OP-GRZ cows had fewer lying bouts than CB-TMR (5.4 vs. 6.5 ± 0.5 bouts; $p < 0.01$) without differing in lying time per bout (55 ± 3 min/bout). No differences were observed in lying behavior between treatments during daylight hours when most of the time all cows were eating TMR. Although, grazing decreased milk yield and lying time, environmental control during supplementation did not have an impact probably because no extreme weather conditions occurred during the measurement period.

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Theatre 4

The effect of mineral-vitamin free choice blocks on transition cows' feeding behavior and health

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Despite the ability to self-select appropriate intakes of specific nutrients is not demonstrated in cattle, providing ad libitum mineral-vitamin (MVit) and energetic blocks may be useful in stressful periods like the transition phase. We assessed the effect of different molasses-based MVit free choice blocks (FC-BLOCK) as alternative to conventional TMR MVit additives on transition cows' feeding behavior and metabolic status. Multiparous Holstein cows ($n=42$), blocked by BCS, parity and previous milk yield, were fed a dry and a fresh TMR at 75% of the MVit requirements. In addition, lactating cows were fed concentrates through robotic milking system. The treatments were: Control (CTL; $n:16$) receiving a top-dressed MVit supplementation to reach 100% of the requirement; mineral-vitamin (MVIT; $n:14$) receiving, in the dry period, a FC-BLOCK followed by a FC-BLOCK with propylene glycol postpartum; linseed (LIN; $n:12$) receiving, in the dry period, a FC-BLOCK with linseed oil and yeast, followed by a FC-BLOCK with propylene glycol postpartum. Feeding and rumination time were recorded daily. Colostrum yield was recorded. Weekly, from -7 to $+20$ DIM, BCS, fecal score (FS) and pH, urine pH (UpH) were recorded; blood NEFA and BHBA levels analyzed. Data were analyzed with PROC MIXED procedures. Treatments had no effect on feeding and rumination time, but a trend for an interaction TRT*DIM was noted with the LIN having lower feeding time (FT) at 5DIM (LIN:163.92 vs MVIT:209.86 and CTL:213.22 min; $P < 0.1$) and higher FT at 20 DIM (LIN:196.73 vs MVIT:191.17 and CTL:157.33 min; $P < 0.1$) compared to the others. First colostrum yield was lower in MVIT compared to CTL and LIN ($P < 0.05$). The treatments didn't affect BCS. At 20DIM, FS and UpH were significantly lower in the LIN compared to the others (LIN:2.65 vs 2.79-MVIT and 3.30 CTL; UpH- LIN:8.03 vs 8.28 and 8.38 for MVIT and CTL; $P < 0.05$). NEFA and BHBA blood levels weren't affected by treatments. The results suggest that LIN FC-BLOCK may play a role in regulating feed intake, but further studies are needed to identify the optimal administration period to avoid intake reduction.

Mineral-vitamin free choice blocks impact on productivity and oxidative status of transition and lactating cows

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Mineral-vitamin (MinVit) and energetic complexes to be offered ad libitum to dairy cattle are proposed as alternatives to conventional TMR MinVit additives for dairy cows. The present study aimed at evaluating the effect of different molasses-based MinVit free choice blocks (FC-BLOCK) in partial substitution to conventional TMR MinVit additives (CTL) on productivity and oxidative status of transition and lactating cows. Three homogeneous groups of cows were fed a basal dry (D) and a postpartum (PP) TMR supplying 75% of the MinVit requirements. The Control group (CTL; n:16) received the remaining 25% as top-dressing on the TMR; the mineral-vitamin (MVT; n:14) group had a FC-BLOCK followed by a FC-BLOCK with propylene glycol during post-partum; and the linseed (LIN; n:12) group had a FC-BLOCK with linseed oil and yeast, followed by a FC-BLOCK with propylene glycol in the two periods. Plasma and milk samples were collected 60 and 7 days before parturition, at parturition (PA) and at 20, 60 and 180 days in milk (DIM), when also milk yield (MY) was measured. MDA, Protein carbonyls (PC), FRAP and ABTS were measured on plasma. Data were analyzed through the univariate procedure of the GLM using values at drying of as covariate. The average MY was similar at 180 DIM. None of the oxidative status markers differed among groups but PC showed highest values at 20 DIM compared to PA and 7 days before PA in the CTL (4.03 vs 3.30 and 3.16). The ABTS tended to have lower values at PA (24.39 on average). Overall, at 20 DIM, ABTS and FRAP appeared negatively correlated with MY ($r=-0.324$; $p=0.033$ and $r=-0.345$; $p=0.025$) and PC positively correlated with MY ($r=0.311$; $p=0.039$) and ECM ($r=-0.354$; $p=0.022$). At the same interval, MDA appeared positively correlated with NEFA ($r=0.513$; $P<0.001$) and BCS loss from parturition ($r=0.300$; $p=0.027$). The use of MinVit free choice blocks is a valid alternative to conventional additives. Oxidative stress is increased by catabolic activities in dairy cattle.

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Theatre 6

Changes in plasma metabolic profile of long-transported Charolais young bulls

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Long transport can cause stress to cattle and thus affect health and growth performance. This study investigated the effects of long transportation from France to Italy on plasma metabolic profile of young beef cattle. Eighty Charolais bulls were randomly divided in 2 groups: Yeast and Control. Yeast group received slow-release nutritional boluses with live yeast strain *S. cerevisiae* CNCM I-1077 and Se-enriched yeast before transportation and were weighed and blood sampled before leaving the commingling center in France (day -1), upon arrival at the fattening unit (day 0), and 7 days after arrival (day 7). Plasma traits included protein, energy, hepato-muscle, and mineral profiles, as well as cortisol and NEFA. Plasma traits and BW were analysed through a repeated linear mixed model with supplementation group, pen nested within supplementation group, time, their interaction, and age of the bull as fixed effects, and bull as random. Transport affected BW, which dropped on day 0 and did not fully recover on day 7. Plasma traits analyses indicated a framework of stress, energy deficit, and muscular damage, with a certain degree of dehydration and hepatic stress due to transport at day 0. Cortisol peaked at day 0 together with other plasma traits, highlighting stressful conditions and physiological responses to recover homeostasis. The supplementation increased plasma Se in the Yeast group, likely due to Se-enriched yeast supplementation. Metabolic profile analysis helps identify stress conditions, enabling preventive actions to facilitate the adaptation of the animals to the receiving fattening unit.

Effects of rumen protected dry grape extract supply on milk performance, body temperature, and breathing behavior of Simmental cows during heat stress

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Heat stress in dairy cows may be successfully faced with the supply of natural antioxidant. This study aimed to evaluate the effects of rumen protected dry grape extract (NorGrape® BPO) supply (rich in water-soluble polyphenols) on milk performance, rectal temperature (RT), and panting response of mid-lactating Simmental cows during heat-stress. Thirty cows (15/group) were blocked by DIM, MY, and parity and randomly assigned to control (CTR) or treated (BPO) group. In the BPO group, 470 mg/d of NorGrape was mixed with corn meal and top-dressed onto TMR for each cow for 35 d during summer. Heavy breathing time (HB) by sensor (Allflex®), MY, and RT data were recorded, and milk samples were weekly collected. Data were analyzed with the PROC GLIMMIX of SAS. Overall, BPO cows had higher MY (28.47 vs. 27.01±1.32 L/d) and yield of protein and casein than CTR (P<0.01). Overall, BPO had lower RT than CTR cows (38.63 vs. 38.74±0.10°C; P<0.01), especially in the morning at 17 (-0.28 °C) and 28 d (-0.32 °C) when THI was 69 and 75, respectively. BPO had lower time spent for HB (61 vs. 104±3 min/d; P=0.05) than CTR group. From 25 to 34 d, when THI increased above 74, the HB in BPO cows was 87 min/d less than CTR (P<0.01). These outcomes suggest that supply of rumen-protected polyphenols from grape extract can help cows to better modulate the thermoregulation during heat stress, which in turn leads to energy being diverted to sustain milk production.

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Theatre 8

Could the inclusion of grape pomace in the diet of dairy ewes in late gestation influence the quality of colostrum and its microbiota?

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This study aimed to test if the inclusion of small amount of grape pomace (GP), a byproduct rich in antioxidant compounds, could improve the quality of colostrum and modify the microbiota. Twenty ewes in late gestation (15 before lambing) were divided in two groups: one group was fed with a basal diet composed by a total mixed ration (CON), whereas the second group was fed a basal diet supplemented by 50 g/d of grape pomace (GP). The ewes received the supplementation of GP until the lambing. The colostrum was collected manually within 12 hours after the lambing and then stored at -80°C for DNA extraction. The inclusion of GP did not affect the fat (9.57 and 10.7%, CON and GP, respectively) and protein concentration (3.8 and 15.6%). The IgG concentration was 54.1 vs 67.0 for CON and GP (P=0.13). The main FA found in the samples were C16:0, followed by C18:1 cis-9 and C14:0. Concerning the taxonomic classification, the colostrum samples from GP had higher abundance of genera *Lawsonibacter*, *Harryflintia*, *Anaerotruncus* and *Monoglobus*, belonging to Ruminococcaceae family, compared to samples from CON. On the contrary, the genera *Butyrivibrio* and *Anaerotignum*, belonging to Lachnospiraceae family, were higher in CON compared to GP. In conclusion, the introduction of 50 g/d of GP in the diet of sheep in late gestation did not evidence any negative effect on quality of colostrum but could modify its microbiota. Acknowledgements: This work was financially supported by NextGenerationEU; PNRR M4C2, CN00000022 AGRITECH.

Application of individual piglet birth weight and within-litter birth weight variation as proxy predictors for pre-weaning survival

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The aim of this study was to evaluate the effects of birth weight (BW0), within-litter birth weight variation (BWvar) and other sow and environmental effects such as parity, sex, litter size, year, season and farm on piglet survival from birth to weaning. The binary classification of survival was used to predict the probability for survival at a given time point using logistic regression models. Next, the optimal cut-off values for BW0 and BWvar individually and as BW0 / BWvar index as predictors for pre-weaning survival were estimated by the Receiver Operating Characteristic (ROC) curves analysis. Data consisted of 66,464 piglet records from 1,661 sows obtained from the FBN and Agroscope research farms. The fixed effects of the sow and environmental effects fitted in the mixed models influenced significantly the piglet survival at birth but their significance changed with age and management interventions such as fostering. The BW0 had the greatest effect on survival. The ROC analysis revealed that piglets below a BW0 cut-off value of 1.18 kg (Area Under Curve, AUC = 0.67) had a lower survival probability at birth than their heavier counterparts. With cut-off values of 0.277 g (AUC = 0.55) and 3.81 (AUC=0.65) for the BWvar and the index, respectively, both did not improve the accuracy of prediction of pre-weaning survival compared to the BW0. In conclusion, individual piglet BW0 was the most important predictor for pre-weaning survival and can be used effectively as single easy-to-measure proxy for pre-weaning survival.

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Theatre 2

Deciphering the Impact of Feeding Behavior on Growth Performance and Nutrient Efficiency in Grower-Finisher Pigs

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In this investigation, we examined how weekly feed intake patterns influence growth performance traits and nutrient efficiency in growing pigs, hypothesizing that feeding behavior variations contribute to individual growth rate and feed efficiency (FE) disparities within a group. Our analysis leveraged a comprehensive dataset capturing growth performance across pigs weighing 27 to 110 kg, including weekly body weight (BW) measurements, average daily gain (ADG), and daily feeding behavior traits monitored by automatic feeders. We calculated the Index of Irregularity of Feed Intake (IIFI) from these data. The IIFI reflects on the relative cumulative feed intake over the relative time in a weekly interval and is calculated from that monotonically increasing step function. Employing multiple regression models and correlation analyses, we elucidated the relationships between feeding behaviors—total visits, feeding duration, visit size, and feeding rate—and weekly growth performance. Notably, strong correlations ($r > 0.70$) emerged between ADG and average daily feed intake (ADFI), alongside moderate correlations ($0.3 < r < 0.7$) for FE with several parameters. Our regression models, significant for both ADG ($R^2 = 0.67$) and FE ($R^2 = 0.37$), underscored the predictive relevance of BW, ADFI, feeding duration, and visit size for ADG, and highlighted IIFI and visit size as significant for FE. A novel finding was the positive correlation between carcass fat content and ADFI, visit size, and IIFI, suggesting that lower IIFI reflects more consistent feeding patterns. Our results affirm feeding pattern traits as reliable indicators of ADG but not FE, with potential implications for predicting body composition at the fattening period's end. PIGWEB received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

Optimizing lactation performance of hyperprolific sows through *Scutellaria baicalensis* supplementation
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Increased sow prolificacy has led to larger litter size, adversely impacting piglet birth weight, vitality, survival, and growth. Parturition and lactation expose sows to risk of inflammation and oxidative stress, hindering milk production and piglet growth. *Scutellaria baicalensis* (SB), an Asian flowering plant with anti-inflammatory and antioxidant properties, could potentially enhance sow health, stimulate mammary gland synthetic capacity, ultimately improving piglet growth. This study investigated the effects of SB supplementation in lactation diet on colostrum and milk production, litter survival and growth. On day 108 of gestation, 24 Danbred sows (2-4 parity) were assigned to control or SB groups. Piglets were individually weighed at birth and weekly during lactation, and sow's milk yield was predicted. Colostrum was sampled at birth; milk samples were collected on day 3, 10, 17, and 24 of lactation. Supplementation of SB did not affect the number of live-born piglets, but increased piglet's birth weight ($P < 0.01$). Litter weight during lactation was greater in SB sows ($P < 0.05$). Milk yield ($P = 0.06$), litter size ($P = 0.08$), and litter survival ($P = 0.10$) tended to be higher in SB supplemented sows. Colostrum protein and solid not fat ($P < 0.01$), and milk fat and dry matter ($P < 0.05$) were lower, while colostrum lactose ($P < 0.01$) was higher in SB sows. In conclusion, SB supplementation improved piglet birth weight and enhanced litter growth during lactation. This suggests SB's potential for improving lactation efficiency in hyperprolific sows, thus studies are needed to understand the underlying mechanisms. Funded by EU's H2020 research and innovation program, PIGWEB grant agreement No 101004770.

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Theatre 4

Piglet response to L-leucine supply during the post-weaning phase depends on piglet birth weight

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This trial aimed to investigate the piglet response to leucine during the post-weaning phase as a function of piglet birth weight. The hypothesis was that leucine improves piglet performance, especially for those with a light body weight. In this trial, 180 piglets weaned at 28d of age were allocated to six treatments for five weeks in a 2×3 factorial design: two body weight categories (BW) depending on the birth weight and three levels of digestible leucine/lysine in the diet (LEU) (deficient: 85%, at the requirement: 100% and in excess: 115%). Growth rate, feed intake and feed conversion ratio (FCR) were recorded and analysed with mixed models, using BW, LEU and their interaction as fixed factors. The BW×LEU interaction was significant for final body weight, growth rate and feed intake ($P < 0.01$). The FCR was not influenced by the interaction, BW or LEU. In the group of light piglets, those fed 100% LEU had significantly higher feed intake, growth rate and final body weight than those fed 85% LEU ($P < 0.05$). In the group of heavy piglets, those fed 115% LEU performed best, with a numerical or significant difference in feed intake, growth rate and final body weight compared to the two other LEU treatments. Diets with 85% LEU appeared deficient for all piglets. The heavy piglets responded positively beyond the level considered to be 100% of the requirement, while for the light piglets, this level seemed optimal. In both cases, the response to leucine seems to be driven by the feed intake. Moreover controlling the Leu level at 100% SID Leu:Lys allowed to homogenise the growth performance of piglets. This work was supported by the PIGWEB project, which has received funding from the European Union's Horizon 2020 programme under grant agreement No 101004770.

Oro-caecal transit time measured with the lactose-¹³C-ureide breath test in pigs fed high and low fiber diets
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The lactose-¹³C-ureide (L¹³CU) breath test measures oro-caecal transit time (OCTT) in humans non-invasively. We used the L¹³CU test in pigs as an alternative to intestinal cannulation. Pairs of male littermates (n=22) were fed low or high dietary fiber (DF) (LF, 2.8% DF; HF 6.5% DF) diets from age day (d) 50. At d75 pigs were injected i.m. with 30 µg/kg body weight lipopolysaccharide (LPS) or NaCl (HF-LPS, HF-NaCl, LF-LPS, LF-NaCl; n=9-10/group). At 12 h post-LPS pigs received 400 mg of unlabeled lactose-ureide, and 12 h later at d76, 400 mg L¹³CU together with 33% of the daily feed amount, followed by 2 x 33% of daily feed allowance 4 and 10 h later. Saliva and breath samples were taken at -30, -15, 60, 90, 120, 150, 180, 210, 240 min, then hourly until 12 and at 24 h post L¹³CU feeding. Breath and saliva ¹³CO₂ enrichments were measured by isotope-ratio mass spectrometry, and OCTT, area under the enrichment-time-curve (AUC), and maximum enrichment (Emax) were calculated. Data were analysed using the PROC GLIMMIX procedure (SAS). In breath, OCTT (3.4 vs 2.9 h) was longer and Emax (7.3 vs 11.9 ‰) lower for LF-LPS vs LF-NaCl pigs (p<0.05). HF-LPS pigs had shorter OCTT (2.9 vs 3.4 h) but greater Emax (9.7 vs 7.3 ‰) compared to LF-LPS pigs (p<0.05). In saliva, AUC of LF-LPS (50.9 vs 34.9 ‰·h; p<0.05) was greater than in LF-NaCl pigs. The L¹³CU breath test successfully measured OCTT in pigs, whereas saliva results were not as sensitive. The HF compared to LF diet shortened OCTT in LPS-treated pigs. Treatment with LPS prolonged OCTT and reduced Emax in LF pigs, suggesting an effect on microbiota. EU Horizon 2020 Research & Innovation programme: PIGWEB Grant # 101004770

Bones and meat quality of pork from early and late immunocastrated male pigs

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Immunocastration is a strategy to reduce boar taint, mainly present in meat from entire males, but it has an effect on pigs body composition. The aim of the project was to characterize the bones and meat quality of pork from entire male (EM) pigs compared to early (EIC) and late immunocastrated (LIC) pigs. For this purpose, (LargeWhite x Landrace) x Pietrain pigs were fed the same diet and randomly distributed in 3 groups. EIC (n=14) received the first Improvac® vaccine (V1) 13 weeks before slaughter (AM) and the second (V2) at 8 weeks AM, while LIC (n=14) received V1 at 8 weeks AM, and V2 at 4 weeks AM. Pigs were slaughtered at 23.4±0.3 weeks of age. Carcasses were scanned with the computed tomography device (Philips Brilliance 16) to determine bone volume and density. Samples of loin for histological, sensory and fat analysis were obtained. No differences (P>0.05) between sex types were found in bone volume, polyunsaturated fatty acids and muscle fiber number. However, proportion of very high density bone was the highest in EIC. Drip loss was higher in EM than in EIC, LIC being in between. Saturated fatty acids in subcutaneous fat were the highest and monounsaturated the lowest in LIC. The proportion of fast-twitch oxidative fibers was higher in EM than in LIC. The boar taint (sensory analysis) was higher in fat from EM than those from the IC pigs. Thus, immunocastration, both, early and late, reduces boar taint and does not significantly affect muscle histological and bone characteristics. This work is part of a TNA within PIGWEB project that has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

The space allowance affects selected pork physicochemical traits of native Polish Pulawska pig breed
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The existing research on commercial pig breeds shows no or limited variation in meat physicochemical attributes caused by space allowance. The goal of our study was to examine the effect of space allowance on the native Pulawska pig. In total 80 fatteners (males and females), were divided into 3 groups: control (CON), with a 1.0 m²/animal; EXP1 – 1.5 m²/animal, and EXP2 – 2.0 m²/ animal. Animals were slaughtered after reaching a weight of 100-110 kg, and selected slaughter traits and meat quality traits (m. longissimus thoracis et lumborum – LTL) were examined on 42 from a total of 80 fatteners (14 per group). The average cold carcass weight was 92.9 kg (± 55.8). Females were characterized by greater leanness (P< 0.001), and by 7.1% heavier loins (P= 0.039) compared to males. The LTL of EXP1 and EXP2 pigs was characterized by a higher pH (5.87 and 5.70 vs 5.65) compared to CON. EXP1 pork had a lower EZ drip loss (1.51% vs 2.81%), and darker colour (L* = 47.7 vs 50.8) compared to CON. The intramuscular fat content did not vary between groups or sexes, and was 1.4 – 2.0%. To conclude, the space allowance in Pulawska pig fatteners significantly affected the physicochemical traits of pork defining its culinary and technological usefulness. Pigs with greater than commercial space allowance, 1.5 m²/pig, characterized with dark-firm-dry abnormal quality attributes. Pigs with a space allowance of 2.0 m²/ animal were characterized by meat attributes located between the other pig groups. This project has received funding from the European Union's Horizon 2020 Research and Innovation Programme under Grant Agreement No 101000344.

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Theatre 8

People want research to improve pig welfare & health, says survey

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An online citizen survey was done by the EU-funded PigWeb research consortium in 2023. The survey was done to improve transparency in the pork industry, address animal welfare concerns in pig farming, and give research partners an insight into people's attitudes toward pork consumption and animal welfare. The survey was divided into three sections. Participants were asked about their attitudes towards meat consumption, their understanding of pig farming methods, and attitudes towards pig production research and animal welfare. 450 EU citizens took part via an online survey, 95% of them consume meat but 9% no pork. Especially interesting for the pig research community are the results regarding the acceptance of the different research purposes and methods. More accepted are purposes which aim to a higher welfare (80%) of the pigs and improve the health (82%) or the quality (74%) of the meat. Less accepted is research to lower the costs (50%) of the breeding. This mirrors also in the methods of research. Highly accepted are modelling (49%) and non-invasive methods (50%). Hardly acceptable for the citizens are methods resulting in discomfort (19%) or even pain (7%) for the pigs. Overall, the data indicates a strong concern for the welfare and well-being of pigs, with experiments causing pain or distress being widely rejected. The data also reflects varying degrees of acceptance for experiments with non-invasive methods and those aimed at reducing pig production's environmental impact and costs. In summary, research practices in pig farming should prioritise ethics, transparency, environmental sustainability and consumer education.

Conceptualisation of “animal discomfort” using the domesticated pig (*Sus scrofa*) as model

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The term “discomfort” is commonly used in biomedical research, animal experimentation for agricultural aims, and animal welfare legislation. Despite its widespread use, the concept of “animal discomfort” remains poorly understood. To address this, we conducted a Walker and Avant concept analysis using the domesticated pig as a model to establish an operational definition of animal discomfort. Using the keywords “ANIMAL” and/or “DISCOMFORT” and/or “DEFIN*” in the Scopus database, 2,594 documents published in English were retrieved. Documents were screened fully following the criteria: 1—a definition and/or measurement of discomfort in animals; 2—definition and/or measurement of pain, suffering, or sickness in pigs. In total, 118 documents were retained for further analysis. Animal discomfort intersects on three domains: physical/sensory, physiological, and mental discomfort. The presence of discomfort leads to a sensation of uneasiness that results in behavioural signs of attempting to avoid or reduce the source(s) of this affective state. At EAAP, we will present in detail: the use of the term “animal discomfort” across the three aforementioned domains; the attributes of “animal discomfort”; a model case in which discomfort occurs; a list of indicators of animal discomfort; and our proposed operational definition. Our work covers a central aspect of animal experimentation and has the potential to improve legislation consistency and animal research integrity as well as ultimately to promote a more sustainable livestock production.

Session 78

Theatre 10

Towards a self-initiated and automated judgement bias task for pigs

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To improve the welfare of farm animals, accurate assessment of their affective state is critical. One approach is to use a judgement bias task (JBT). However, JBTs have limitations like lengthy training and occasional lapses of motivation. To address these issues, we developed a self-initiated, automated JBT for pigs as part of the PIGWEB project funded by the European Union’s Horizon 2020 research and innovation programme under grant agreement No 101004770. Pigs could touch a screen to initiate a trial, followed by hearing one of two tones indicating either that they could collect a food reward from a feeder (go) or stay away from the feeder to avoid an air puff (no-go). Of the 23 pigs, 18 were successfully trained. Judgement bias was measured by the pigs’ responses to two ambiguous tones. As an affect manipulation, pigs were either moved to barren (3.4m²) or enriched (11.2m²) pens. During 13-min test sessions, pigs initiated 30-150 trials. Preliminary analysis with mixed models showed that pigs discriminated between the reference and ambiguous tones, demonstrating a monotonically graded response. However, our prediction that barren housed pigs would show more no-go (‘pessimistic’) responses to the ambiguous tones was not supported, indicating no significant difference in affective state. In conclusion, pigs can learn this self-initiated and automated JBT well, but more research is needed to understand why the affect manipulation did not have the expected effect.

State-of-the-art heart rate sensors for grouped-housed pigs

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A literature and market study was performed in the PIGWEB project (H2020, 101004770) to search for minimally invasive and practical heart rate sensors for pigs. Sensors designed for other livestock, dogs and humans were also considered. Several sensors were found, incl. electrocardiographic sensors, photoplethysmographic sensors (PPG), and ballistocardiography and seismocardiographic measurements. In addition to contact sensors, different wavelengths can be captured with cameras such as ultrasound, RGB, near-infrared, but also thermal, laser, non-contact shortwave microwave radar system and non-contact magnetocardiography. These sensors measure mechanical effects (body surface displacement, superficial perfusion, intrathoracic dynamics), thermal effects (blood flow-induced temperature variation) and bioelectrical effects. Several challenges still remain with the available sensors. Validation is often only performed in controlled environments and fixated animals. Some of the less invasive techniques still require impractical measures (shaving skin, applying gel, etc.). In the long term, pigs could lose or destroy the sensors. Limited battery life is also an issue. Video measurements require no contact with the animals, but often also require fixation due to heavy motion artefacts. According to the literature, the most promising techniques are PPG sensors combined with motion sensors to filter out motion artefacts, which can be applied using ear tags. Advanced video processing could also be an option, where pigs are individually evaluated on specific body parts while resting. No readily commercially available, practical, tested, and proven solutions were found that could be used in group-housed pigs of various ages. More work is thus needed to bring these techniques to a higher technology readiness level.

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Theatre 1

From pen to pasture: customizing welfare protocols for small ruminants in different production systems

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In animal welfare research, a distinctive and insightful perspective is emerging through applying a "bottom-up approach" in the study of small ruminants. Traditionally, welfare assessments have often been top-down, relying on standardized indicators and generalized guidelines. However, the bottom-up approach recognizes small ruminants' unique needs and behaviours, such as sheep and goats, at the individual and flock levels. This study emphasizes tailoring welfare assessment protocols for small ruminants in different production systems. The focus is developing practical and reliable on-farm tools that assess and address welfare concerns. Research has shown success with protocols combining animal-based indicators with husbandry assessments. Existing protocols like AWIN provide a foundation for development in different systems. These protocols have been adapted for semi-extensive farming conditions and demonstrate good feasibility across farm sizes. However, many aspects of grazing and extensive systems still need to be more adaptable. To close this gap, the research presented will focus on the welfare perception by farmers specifically on native breeds and the necessary commitments to propose a suitable welfare protocol adapted to their reality. This work was supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Effect of suckling method on the response of lambs to social isolation and weaning

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The objective was to test whether the sucking system (natural vs. artificial) would affect lambs' response to social isolation and weaning stress. Lacaune lambs (n = 80) were divided at birth into 2 balanced groups according to birth weight, sex, and lambing type (single or multiple). Treatments were: 1) natural suckling (NAT; n = 40), where lambs were kept with their mothers from birth until weaning at week 5 of age, with no human contact, and 2) artificial suckling (ART; n = 40), where lambs were fed milk replacer ad libitum. At weaning, an arena test was carried out by placing lambs in the arena for 8 min and recording the behavior using a camera (number of squares crossed, frequency of jumping and sniffing, vocalizations, and time spent moving). Blood samples were collected at days 0, 1 and 2 relative to weaning for the analysis of cortisol (CORT), haptoglobin (HP), lactate dehydrogenase (LDH), creatine kinase (CK), and free fatty acids (FFA). In the arena test, NAT lambs sniffed more (P < 0.01) than ART, but ART lambs had a greater (P < 0.02) number of walls jumps than NAT. Additionally, ART lambs tended (P < 0.06) to vocalize more than NAT. During the 2 d post-weaning, ART lambs had lower serum values of CORT (-27%; P < 0.10), LDH (-43%; P < 0.05), CK (-97%; P < 0.001), and FFA (-26%; P < 0.01) than NAT lambs. Weaning increased (P < 0.01) Hp at day 0 (0.30 mg/mL), day 1 (0.45 mg/mL), and day 2 (0.66 mg/dL) in both NAT and ART lambs. In conclusion, The ART lambs showed more active behavior in the arena test and experienced less weaning stress than NAT lambs. Acknowledgements: funded by the Ministry of Science & Innovation (Project RTA #PID2020-113913RR) and TechCare (Contract # 862050) H2020 UE Program.

Session 79

Theatre 3

Supporting breeders to better manage animal pain: the case of sheep tail docking

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Pain management during preventive interventions on sheep farms will be increasingly regulated in Europe in the coming years. The aim of this study was to assess the pain generated by docking practices used on sheep in France, and the effectiveness of an analgesic protocol that could be implemented by farmers. Behavioral, physiological and zootechnical indicators were studied 1) in March 2022 on 48 2-day-old lambs (16 control lambs (T), 32 lambs with rubber ring docking without analgesia (16 E) and with analgesia (16 EA)) and 2) in March 2023, on 80 13-day-old lambs (16 control lambs (T), 32 lambs with rubber ring docking : without analgesia (16 E) and with analgesia (16 EA) 32 lambs with hemostatic clamp: without analgesia (16 H) and with analgesia (16 HA)). An analysis of variance (ANOVA) test was used to compare the results of lambs of both ages having undergone rubber ring docking and the two docking methods in 13-day-old lambs. Rubber-ring docking at 2 days of age elicited very few pain-related behaviors. The 13-day caudectomy methods (rubber ring and forceps) induced acute pain in the lambs (elevated serum cortisol levels of 127 ± 58 nmol/L for E and 132 ± 104 for H, 30 min after caudectomy), but with different behavioral characteristics: increase in the number of events for rubber ring caudectomy (255 ± 68 for E and 208 ± 62 for EA vs. 29 ± 7 for T), and appearance of tremors associated with stooped posture, with no visible increase in the number of events for forceps caudectomy (30% of time spent in this position over 2 hours after caudectomy for H and 19% for HA vs. 0% for T). This method resulted in hyperalgesia in most lambs (assessed by Von Frey filaments), persisting for at least 6 days (on average 20% of sensitive lambs for E and EA and 62% for H and 73% for HA). The analgesic protocol tested showed a visible effect only in 2-day-old lambs. However, local anaesthesia is proving difficult for farmers to apply in the field. Further work is needed to find an effective analgesic protocol that can be applied by farmers. Project funded by Interbev

Ovine lameness in Ireland – An assessment of farmer management practices via survey

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Currently, a paucity of information exists as to the farm based management factors that are used to detect and control cases of ovine lameness within Irish farms. The objective of this study was to investigate these farm based management factors through the distribution of an online and in-person based survey between January 2023 and March 2024. In total 205 lowland Irish sheep farmers responded to the survey. The mean flock and farm size reported by respondents was 201 (SD = 184.9) ewes and 54 (SD = 49.7) hectares. In order to identify lameness, observing sheep grazing was the main detection method reported by 60.9% (95% CI: 55.3, 66.5) of respondents. Once a lame sheep was detected, 64.9% (95% CI: 58.3, 71.4), of farmers reported to treat the animal within 1 week, while 83.6% (95% CI: 78.5, 88.7) of respondents reported ‘turning over’ the lame sheep to inspect the hoof prior to treatment. As a control method for lameness, foot bathing was not carried out by 17.2% of respondents (95% CI: 11.6, 22.9). Of those that did footbath 33.1% (95% CI: 25.2, 41.0) didn’t know to what concentration their footbath solution was made up to and 8.4% (95% CI: 3.8, 12.9) of respondent’s released sheep back to the field immediately after foot bathing. Routine foot trimming was carried out by 52.6% (95% CI: 45.1, 60.1) of respondents with 28.3% (95% CI: 19.1, 37.5) of these respondents trimming to treat cases of infectious lameness and 21.7% (95% CI: 13.3, 30.1) trimming as a preventative foot-care measure. This study demonstrates the variation in lameness management practices, further work is required to fully understand and disseminate best practice to flock-keepers.

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Theatre 5

The impact of lameness on the body condition score of ewes on three Irish farms

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Currently, there is a paucity of information on the impacts of lameness have on the farm production targets of Irish sheep farms. The objective of this study was to investigate the level of persistently lame sheep in commercial flocks and to determine the effect in which lameness had on ewe BCS from mating to mid-pregnancy. Three flocks with a mean flock size of 259 ewes (SD=69.5) were visited twice over a four month period and lame sheep (Locomotion score ≥ 1) were identified prior to mating and again during mid-pregnancy when ewes were ultrasound scanned to determine pregnancy status. All flocks were grazed outdoors on perennial ryegrass based swards and brassica crops sown into cereal ground post-harvest from mating to mid-pregnancy. Persistently lame sheep were classified as sheep lame at the mating visit that also presented as lame again at mid-pregnancy. Lameness prevalence identified across farms A, B and C were 11.6%, 13.7% and 6.5% at mating and 12.5%, 15.1% and 20.1% respectively at mid-pregnancy. On farms A, B and C, 16.7%, 28.0% and 17.4% of lame sheep were identified as persistently lame. Across the three farms ewe BCS change between mating and mid-pregnancy was not effected by lameness status at the time of BCS recording ($P > 0.05$). Farm was a significant factor in the model ($P < 0.05$), the BCS of ewes in the flock with the highest proportion of thin ewes (percentage ewes BCS < 3.0) at mating was effected by lameness ($P < 0.05$). The results of this study show that a proportion of lame sheep will continue to have lameness issues despite initial intervention. Furthermore, while overall BCS change was not effected by lameness, the effect varied between farms indicating that lameness severity, treatment strategy and nutritional management may all have an impact.

Relationships between on-farm animal welfare assessment and milk yield in dairy sheep farms

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In Italy, due to the increasing public awareness for high standards of welfare and quality of animal production, a method to assess animal welfare was implemented by using an on-farm checklist called "Classyfarm". The aim of the present study was to evaluate if the animal-based measures obtained with Classyfarm checklist, were associated to milk yield in a cohort of dairy sheep farms (DSF) located in Tuscany region (Italy). A total of 47 DSF were included. Six different areas, with the related items, were scored: biosecurity (15 items), management (16 items), structure (14 items), animal-based measures (ABM) (10 items), hazards and risks (HR) (8 items) and total welfare (TW) (63 items). Data about daily milk yield produced at the time of Classyfarm assessment were recorded from each DSF. A regression tree model was used to evaluate possible milk yield cut off value for the different Classyfarm areas. Results showed no differences for biosecurity, structure, ABM, HR and TW area while significant difference was found for management area. DSF with management area score > 65.6% showed higher milk production compared with others (1.57 vs 1.05 L/sheep). Our results showed that high management score (e.g. knowledge of the shepherd, ratio shepherd to flock size ratio, flock separated in different groups according to physiologic status, treatment of sick animals, type of feed, cleanliness of drinkers and floors) could have a relation with animal productivity. In conclusion, the application of Classyfarm checklist may enhance farmers' understanding of the animal welfare, leading to an improvement of animal health and productivity.

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Theatre 7

Tree shade reduces heat stress in pregnant ewes

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This study aimed to evaluate the effect of access to tree shade on heat stress indicator traits in ewes during late pregnancy. Ambient temperature (AT) and relative humidity (RH) were used to obtain a temperature-humidity index (THI). Dormer and South African Mutton Merino (SAMM) ewes (166 in total) were randomly allocated to 9 paddocks with access to tree shade (SH) and 9 paddocks without access to shade (NS), 14 days before the commencement of lambing. The respiratory rate (RR) and rectal temperature (RT) of each ewe were assessed between 12h00 and 14h00 over 3 random days. On the 14th day postpartum, ewes were removed from the treatments and wool samples were shorn from each ewe. The wool samples were cleaned, cut into 1 to 3 mm pieces using surgical scissors, and mixed with methanol for cortisol extraction. The extracts were analysed for cortisol concentrations using DetectX® Cortisol Enzyme Immunoassay Kits. Wool cortisol concentrations, RR and RT were subjected to linear mixed models involving the fixed effects of breed, shade treatment and ewe age as well as THI as a covariate in ASReml. Shade treatment affected both RR and RT ($p < 0.001$) and interacted with THI ($p < 0.001$). Animals in SH paddocks had a lower RR and RT, and the treatment effect was accentuated at higher THIs. Dormer ewes had a higher RR than the SAMM ewes ($p < 0.05$). Both traits were repeatable, at 0.32 ± 0.08 for RT and 0.16 ± 0.08 for RR. The cortisol levels in barren ewes were lower ($P < 0.01$) than in those that lambed. Shaded ewes tended to have lower wool cortisol concentrations than NS ewes ($P = 0.057$). Tree shade reduced the effects of heat stress in pregnant ewes and benefitted sheep welfare.

Human perception of the vocal expression of the emotions in goats

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The ability to recognize animal emotions is fundamental to promote their welfare. To investigate the human ability to interpret goats' vocalizations, two online surveys were prepared: one for people with no frequent direct contacts with goats (NFC) and one for people with frequent direct contacts with goats (FC). Respondents were asked to listen to four bleats and associate them to different contexts of emission potentially related to different underlying emotions (i.e., waiting for feed, pre-partum labour, mother-kid reunion, mother-kid separation) and to describe them by using qualitative descriptors (i.e., aggressive, active, curious, frustrated, irritated, relaxed, satisfied, and suffering). Two hundred and thirty-nine NFC surveys and 81 FC surveys were collected. Female respondents showed a higher ability to recognize bleats associated with pre-partum labor than males (81.7% vs 54.5%; $P < 0.05$); the ability to better recognize emotions conveyed by animals' vocalizations by female gender has been highlighted in previous studies, and could be attributed to its greater level of empathy. Additionally, FC people outperformed NFC people in identifying most emission contexts (waiting for feed: 56.8% vs 43.5%, $P < 0.05$; pre-partum labour: 86.4% vs 74.5%, $P < 0.05$; mother-kid separation: 72.8% vs 52.3%, $P < 0.001$). Principal Component Analysis highlighted a trend of negative contexts (mother-kid separation and pre-partum labour) to separate from the others, based on qualitative descriptors. For FC people, this was related to higher values of suffering and lower values of positive descriptors, whereas for NFC people, it unexpectedly correlated with higher relaxed and satisfied values and lower negative descriptors values. Our findings are in line with those of previous studies suggesting that gender and experience influence humans' ability to interpret goats' vocalizations.

Session 79

Theatre 9

Is welfare of sheep kept in marginal areas influenced by seasonal climatic condition?

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Sheep are commonly considered well adapted to be farmed in marginal areas. From an environmental perspective, extensive farming in these rural areas can lead to a more sustainable farming system, increasingly valued also by consumers. However, pasture productivity, the primary resource for animal feeding, as well as the availability of resources such as shade and water, is dependent on climatic condition. The aim of this study was to describe how seasonal climatic condition changes affect the welfare of sheep farmed in marginal areas. The welfare of 6 flocks raised in marginal areas was monitored with the AWIN welfare assessment protocol adapted for extensive systems. A total of 72 sheep (12 for each flock) were individually monitored using 24 animal-based measures during two visits, one in summer and one in winter. The proportion of each score of animal-based indicators was calculated to identify possible welfare issues. Good feeding was not influenced by climatic condition, with 50% of the assessed sheep enjoying a BCS=3 in both seasons. In winter, 78.5% of sheep showed a dirty and wet fleece and 62.8% had hooves overgrowth, while in summer the prevalence was 38.8% and 11.1%, respectively, meaning that climatic conditions as well as seasonal management (e.g. trimming, shearing) can affect sheep welfare. In summer, over 25% of sheep had eyes or muzzle lesions, while in winter the prevalence was less than 6%. These preliminary results reveal differences in the welfare condition of sheep raised in marginal areas, highlighting the importance of identifying possible risk factors associated with climatic and environmental conditions.

Development of an operational tool for assessing and managing the welfare of sheep and goats on farms in France

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Animal welfare is a social concern shared by all the small ruminant sectors in France and a priority in their action plans. No consensual tool for assessing and managing the welfare of these animals, adapted to French farming systems, was available. The CMOUBIENE project aims to develop a welfare assessment and management tool for the sheep (milk and meat) and goat sectors, as part of a co-construction approach with all the stakeholders. It is above all the methodology used to build this tool that will be presented here. The tool's welfare indicators (for young and adult animals) have been scientifically validated and selected by the stakeholders. Thresholds were then set for each indicator via an online Delphi survey of a large panel of professionals (sheep and goat farmers, technicians, veterinarians, researchers...). Four classes were selected for each indicator: excellent, good, intermediate and unclassified. The tool was implemented on Excel for sheep and on a smartphone application for goats. A training kit was developed, and 24 technicians were trained to test the tool in 30 farms in each sector. According to the results of these tests (not yet available in September 2024), and the feedback from technicians and farmers, tools, training kit and thresholds may be reviewed/adapted if necessary. The results obtained will be presented not only by indicators, but also by criteria (following expert workshops) and principles. From 2025, these tools will be deployed on French farms by the sheep and goat sectors. Project funded by CASDAR, Interbev, ANICAP and France Brebis Laitières

Session 79

Poster 11

On farm animal welfare self-assessment tool for dairy goats

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Dairy goat farms have experienced a strong process of intensification in Spain in the last decade, which, in many aspects, could compromise optimal animal welfare (AW). The objective of this work was to develop and implement an AW assessment tool for dairy goat systems in collaboration with relevant stakeholders in the sector. The study had three phases: i) first, we discussed with stakeholders the specific issues they found more relevant to cover in modern dairy goat farms plus a literature review of works that addressed similar objectives; ii) then, a draft questionnaire was developed and discussed with stakeholders; and iii) finally, a selection of farms was made to test the tool. The stakeholders involved included veterinarians, nutritionists, breeders and managers in the dairy goats sector. The questionnaire (Google Form) was organized into the four main domains considered in the Welfare Quality® protocol: good feeding, good housing, appropriate behavior, and good health, including respectively 19, 23, 14, and 3 specific questions. Most of them are answered using a 1-10 scoring system, with specific guidance for score allocation. Twelve farms using a representation of the main feeding systems were used to run an initial test of the tool. The results showed that the categories that exhibit lower average scores and larger differences across farms were: water availability (5.33±2.88) and management (4.88±1.64), density of animals indoors (6.20±2.86), thermal insulation of buildings (5.67±2.07) and hoof care (5.50±1.51). The rest of parameters averaged above 7-8 with very low variation across farms and no differences between the main feeding systems. The questionnaire showed potential to be used as a tool to evaluate the animal welfare status in dairy goats farms and identify 'hot spots' for future actions. Acknowledgement:H2020-PATHWAYS project (101000395).

Effects of drinking diluted seawater on behavioral and physiological responses in crossbred dairy goats under tropical condition

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The high salinity of drinking water is a significant problem in the Mekong River Delta. The present experiment aimed to investigate the effects of various levels of diluted seawater (SW) in drinking water on behavioral and physiological responses, in crossbred dairy goats. The experiment was conducted with four groups (n=5 each) categorized by body weight and milk yield, which included a control group of goats drinking fresh water (SW0.0). The other groups consisted of goats drinking seawater with concentrations of 0.5% (SW0.5), 1.0% (SW1.0), and 1.5% (SW1.5). The experiment spanned 56 days, with the initial 7 days of the pretreatment period and the subsequent 49 days (1st-7th week) as the treatment period. During the experimental period, SWs significantly altered daily water intake (WI) from the second to seventh weeks ($P < 0.05$). Goats from SW1.0 had the highest daily water intake on average, while those from SW1.5 had the lowest. However, dry matter intake was not influenced by salinity ($P > 0.05$). The body water balance was lower, and the plasma ADH level was higher in SW1.5 compared to the other treatments. Additionally, the rectal temperature and respiration rate were higher during 15:00 to 17:00 in SW1.5. However, the plasma electrolyte, creatinine, and HSP70 concentrations did not differ among treatments ($P > 0.05$). The urinary excretion of Na^+ from SW1.5 and K^+ and Cl^- from SW1.0 was higher than those from SW0.0 and SW0.5 ($P < 0.01$). We concluded that crossbred lactating goats adapted to SW0.5 and SW1.0 by increasing WI, urine volume, and subsequently had higher urinary electrolyte excretion. On the other hand, animals responded to SW1.5 by either decreasing WI, altering drinking patterns, and increasing urinary electrolyte excretion.

Session 79

Poster 13

The influence of shearing on the growth performance of feedlot lambs under high temperatures

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With the forecasted increase in temperatures across southern Africa, livestock producers will face greater challenges related to heat stress while aiming to enhance production efficiency. Shearing emerges as a potential solution to address these challenges. To assess the impact of shearing on the production efficiency and heat tolerance of Dohne Merino lambs in feedlot conditions, fifty-six ram lambs were randomly assigned to either be sheared or unshorn and were provided with either a high (11.06 MJ/kg ME) or low (9.29 MJ/kg ME) energy finishing diet. The lambs were fed for 42 days during the summer period ($29^\circ\text{C} \pm 4$) and subsequently slaughtered. Weekly measurements of individual weight, fat depth, and feed intake were recorded, along with respiration rate, surface temperature, and rectal temperature. Shearing did not significantly affect mean weight gain (311 g/day vs. 347 g/day), daily feed intake (1.47 kg vs 1.45 kg), or feed conversion (7.79 vs. 5.98) over the entire growth period ($P > 0.05$). Feed intake was higher in the low energy group ($P = 0.010$), but growth and feed conversion remained unaffected by diet. Unshorn lambs exhibited higher average respiration rates (160 breaths/minute vs. 142 breaths/minute; $P < 0.001$) compared to shorn lambs but showed similar surface temperatures (38.35°C vs. 38.09°C ; $P = 0.095$) and identical rectal temperatures, indicating that increased respiration sufficiently regulated thermoregulation to prevent heat stress from occurring more frequently than in shorn animals. Consequently, shearing of feedlot animals yielded no production benefits for producers and only marginally improved thermal tolerance. Top of Form

Parity, lamb weight, and litter size influence the delivery stance of ewes: standing or lying

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Lambs born to ewes that remain standing at lambing are predisposed to greater birth injuries than those born to ewes that lie down during the process. Consequently, the type of birth (standing or lying) may influence the vigour of the lamb and compromise its survival. This study investigated how factors such as ewe's age, body weight (BW), body condition score (BCS), duration of parturition, litter size, lamb's weight and sex, influence the occurrence of standing vs lying births. One hundred and forty-eight Corriedale ewes (28 primiparous (P): 2-year-old, BW 48.6 ± 4.6 kg, BCS 3.5 ± 0.5, carrying singleton; and 120 multiparous (M): 3-6-year-old, BW 58.6 ± 6.9 kg, BCS 3.5 ± 0.5 carrying both singleton and twins) were monitored at an experimental farm in Uruguay (32°S, 54°W). Data were analysed using GENMOD procedure of SAS. Results are expressed as frequency. For the comparison between P and M, only singleton-lambing ewes were used. Primiparous ewes had more standing births compared to M (28.5 % vs 16 %), and the lowest percentage was registered in 6-year-old ewes (0 %, P<0.001). Litter size influenced the type of birth, with twin births having a higher likelihood of occurring with the ewe standing (34.4 % vs 17.3 %), evident during the delivery of the second twin compared to the first one (21.8 % vs 10.9 %). The weight of the lamb was found to correlate with the type of birth: lighter lambs demonstrated a higher occurrence of standing births, with the percentage decreasing as weight increased (P<0.001). Duration of parturition, maternal BCS, and sex of the lamb, did not affect the type of birth. In primiparous as well as in twin-bearing ewes, lambing without recumbency can be a contributing factor to birth injuries in these susceptible groups.

Animal welfare and microbiological meat safety at religious and conventional slaughtering: a comparative study

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Animal welfare and microbial contamination of carcasses during religious (Kosher) and conventional slaughter were evaluated in cattle and sheep, checking if the DIALREL EU project recommendations are followed. Aerobic colony counts were variable, with ritual slaughter carcasses less contaminated on average than conventional in cattle (n= 80; 623 vs 2,953 CFU/cm²), vice versa in sheep (n=80; 3,434 vs 2,152 CFU/cm²), without statistical significance. Enterobacteriaceae in ritual slaughter of sheep were lower than conventional (17 vs 68 CFU/cm²; P<0.05). E. coli STEC was frequent in both species (52.5% in cattle, 96.2% in sheep), ritual slaughter showing less contamination with major serogroups (P<0.05). Campylobacter and Rotavirus were in fecal samples from both types. Animal welfare critical points were identified as i) containment, ii) cutting of the major throat vessels, and iii) bleeding. Regular checks on the unconsciousness of animals were not carried out in both slaughterhouses. Time to disappearance of signs of consciousness after throat cutting was quite long particularly in cattle (60 to 277 seconds). Properly conducted religious slaughter seems to not significantly impact the microbiological quality compared to conventional. Bovines are at high risk of prolonged suffering during religious slaughter, as indicated by unacceptably long intervals between slaughter and loss of consciousness in some cases. The possibility of applying stunning in cases of prolonged animal agony needs consideration.

Reducing Methane Emissions: A Win-win Solution for Climate and Livestock Agrifood Systems

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Global livestock supply chains contributed about 6.2 Gt CO₂-eq of greenhouse gas emissions (GHG), equivalent to circa 12% of human-induced emissions in 2015. About 61% of these emissions take place on-farm and are dominated by methane (CH₄) from enteric fermentation (75%) and anaerobic digestion of manure (13%). In line with the Paris Agreement, reducing CH₄ and other GHG emissions is an integral part of the strategies aimed at limiting the global temperature increase to well below 2 °C and preferably to 1.5 °C above the pre-industrial level. It also aligns well with the call to cut CH₄ emissions by 30 % by 2030 from 2020 levels of the Global Methane Pledge. This paper aims to provide insights into existing technologies to reduce CH₄ emissions on farms based on animal breeding and management, feed management, diet formulation and precision feeding, forage type and rumen manipulation. Some of these technologies decrease absolute emissions, some decrease emissions yield and others decrease emissions intensity. Further solutions are also provided to reduce emissions during manure management systems. It outlines the readiness of different technical solutions, scalability and their adoption potential by farmers across different livestock systems globally. It discusses the challenges and opportunities for their adoption including the need for climate finance and further research to achieve their wide adoption. This paper builds on recent FAO reports on "Methane emissions in livestock and rice systems: Sources, quantification, mitigation and metrics" and "Pathways towards lower emissions: a global assessment of the greenhouse gas emissions and mitigation options from livestock agrifood systems".

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The integration of manure, soil, diet and animal management to reduce GHG emissions on farm

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Greenhouse gas (GHG) emission mitigation on farms can encompass manure, soil, diet and animal management. All of these components of farm management must be considered prior to changing one aspect to reduce emissions so reductions in one area do not contribute to increased emissions in another. Additionally, extensive (pastoral) production systems can be more difficult to find methods by which methane and nitrous oxide can be reduced than intensive systems in which sources are more controlled. Nonetheless, extensive systems comprise the greatest numbers of ruminant animals across the globe, 88%, and would provide the greatest impact should reduction strategies become available. Current strategies for enteric emissions are fundamentally indirect reduction options. Some of these include supplementation to enhance animal performance, pasture management strategies, genetic selection for efficient and low GHG-producing animals and precision feeding. Manure management strategies to reduce GHG emissions are an active area of research and should become an area where it is possible for farms to produce income and potentially have direct emissions reductions. These strategies include biogasifiers that can produce biofuels, biofiltration systems to compost and clean water for reuse, composting with and without covers and injection of liquid manure into the soil. The attention to GHG reductions in animal production systems should result in a suite of options for producers to choose between to enhance the sustainability of food production.

New York dairy greenhouse gas mitigation: progress made and future potential

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With more than 620000 dairy cows on almost 2800 farms producing more than 7 million Mg of milk per year, New York (NY) is the fifth largest dairy-producing state in the United States. Many NY dairy farms implement beneficial management practices (BMPs) relating to milk and crop production efficiency, nutrient management and soil health. Using Cool Farm Tool (CFT), the mitigation impact of these BMPs on greenhouse gas emission intensity (GHGei), and the potential opportunity to further mitigate GHGei was evaluated for 36 large (>350 cows), commercial NY dairy farms in the 2022 calendar year. Farm size ranged from 350 to 6000 head of predominantly Holstein cows with animal densities between 1.76 and 4.85 animal units/ha, average fat and protein corrected milk (FPCM) yield of 12.7 Mg FPCM/cow/year and an average of 63% homegrown feed. Total FPCM production was 870000 Mg, representing approximately 12% of total NY milk production in 2022. The 2022 GHGei with BMPs currently implemented on farms ranged from 0.63 to 1.06 kg carbon dioxide equivalents (CO₂eq)/kg FPCM (weighted GHGei = 0.81 kg CO₂eq/kg FPCM). A scenario where no BMPs were implemented resulted in a weighted GHGei of 1.17 kg CO₂eq/kg FPCM. A scenario with 100% uptake of BMPs that are both feasible for NY dairy farms and can be modelled through CFT resulted in a potential weighted GHGei of 0.58 kg CO₂eq/kg FPCM. These results show that BMPs already implemented on these NY dairy farms have reduced the weighted GHGei by approximately 30%, with a potential to reduce weighted GHGei a further 20% from the scenario without BMPs. However, characteristics specific to individual farms including infrastructure and economics could limit this full mitigation potential from being achieved. Conversely, as new technologies become available and CFT modelling capabilities develop, additional mitigation opportunities could arise. Furthermore, these BMP scenarios may not be feasible on smaller NY dairy farms with less intensive milk production. Further evaluation of diverse dairy farm types is needed to fully understand the mitigation potential of the NY dairy industry as a whole.

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Trade-offs between methane and ammonia emissions after nutritional interventions in dairy cows

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The complex biological processes and range of gaseous emissions in agricultural systems means there is a risk that mitigation measures implemented to target one gas could lead to increases in emissions of other gases. Understanding these trade-offs will lead to more effective policies and better cohesion across different policy objectives. This meta-analysis included papers where nutritional interventions to reduce methane (CH₄), e.g. feed additive, fibre or lipid content, or ammonia (NH₃), e.g. protein content) were considered. Papers were included if both CH₄ and NH₃ were measured from individual dairy cows after nutritional intervention, or if emissions could be estimated from proxy measures (rumen fluid volatile fatty acid composition for CH₄ and milk urea nitrogen for NH₃). A total of 45 papers were included in the analysis. To allow comparisons to be made between studies, log response ratios (RR, log of mean treatment value divided by mean control value) were calculated for emissions rate (g/day) and emissions yield (g/kg dry matter intake) of each gas to give dimensionless effect sizes. Generalised linear models were created for CH₄ rate RR, CH₄ yield RR, NH₃ rate RR, and NH₃ yield RR. Model terms included geographical region, treatment type, experimental design (continuous or cross-over), whether the gas was measured or estimated, and paper as a random factor. The model means showed small but overall reductions in both emissions rate (-0.01 for both gases), and emissions yield (CH₄: -0.08, NH₃: -0.03) resulting in a win-win situation. Further analysis is underway exploring the data using Bayesian statistics and a further meta-analysis is underway considering trade-offs between CH₄ and NH₃ emissions from slurry stores and the impact of different slurry store cover types.

Milk mid-infrared prediction of methane emission using handheld laser methane detector in smallholders' dairy cattle

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Selection for economically important traits requires thousands of individuals to be recorded, particularly measuring methane emission is expensive, especially in smallholder dairy farming systems. Therefore, the combined use of a handheld laser methane detector (LMD) and milk mid-infrared spectrometry (MIR) are economical means for this purpose. A total of 861 LMD measurements with 3-5 min length each and the corresponding milk MIR profiles were sampled at the ± 3 days from 318 animals of different ages, parity, lactation stages, and breeds in 26 smallholder farms in Ethiopia from July to December 2024 was used. Single variate model using an average of a whole LMD profile and a bivariate modelling by splitting the LMD profile into a mixture of two normal distributions using a standard EM algorithm representing the respiration and eructation status of cows were compared. Partial least square analysis was performed to predict methane emission using MIR data. One-fifth of the data were sampled randomly as a validation set and the rest were used to train the model for prediction. One hundred samplings and predictions were performed and the correlation between predicted and actual measurements was calculated as the accuracy of prediction. Accuracy was 0.29 ranging from 0.06 to 0.44 in the single variate analysis. This improved significantly to 0.68 ranging from 0.56 to 0.8, using bivariate analysis. The improvement resulted mainly from considering the detailed methane production (respiration and eructation) in the prediction although doubling the data may be influential. Results showed that with cost-effective measurements using LMD and help from MIR data, selection for methane emission is feasible in the smallholders' dairy farming system.

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Assessment of methane emission on a Simmental dairy farm

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Among the various greenhouse gases, methane certainly plays an important key role in the environmental impact of a dairy farm. The aim of the study is to assess the methane emissions (ME) of Simmental dairy cows of different parity also considering estimation equations based on the fatty acid profile (FA) of fat and protein corrected milk (FPCM). The equation used for the study is $CH_4 \text{ (g/kg of FPCM)} = 21.13 - 1.38 \times C4:0 + 8.53 \times C16:0iso - 0.22 \times C18:1c9 - 0.59 \times C18:1t10 + t11$. Seven primiparous and 7 multiparous mid-lactation dairy cows with similar milk yields (28 ± 0.7 kg/d) and BCS (3.7 ± 0.1 kg/d) were considered. The methane concentration in breath was measured at feeding twice a day for three consecutive days with the LaserMethaneSmart for 5 minutes at 1 m distance. The average values per animal were analyzed. The third day milk samples were taken to determine FA. Statistical analysis considered one-way ANOVA, Pearson correlation and Bland-Altman analysis. The informative FA for ME, C4:0 ($4.3 \pm 0.07\%$), C16:0iso ($0.3 \pm 0.02\%$), C18:1t10+11 ($1.6 \pm 0.10\%$), C18:1c9 ($18.9 \pm 0.61\%$) were similar between groups ($P > 0.05$) as well as the average ME per cow (318.8 ± 11.33 g/d). The estimated ME was 427.9 ± 22.61 g/d. The measured and estimated ME were not correlated ($r = 0.24$; $P > 0.05$). The mean difference between estimated and measured ME was 109 g/d (CI, 275; -57 g/d) with estimated ME having lower average values than measured ME for low levels of emissions, the opposite for high levels of emissions. This indicates that the relationship between measured and estimated emissions should be further investigated to have definitive results. Work funded by Agritech National Research Center and received funding from the European Union Next-GenerationEU and with PNRR M4C2, DM352/2022 and Tenuta Marianis farms.

Using GWP* to assess the livestock emission impact: the Italian case study

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The global warming potential (GWP) has been adopted to compare the effect of different greenhouse gases (GHG) and find shared strategies to keep the temperature rising beyond 1.5°C. However, this metric does not meet the need to understand the real effect of each GHG on the planet's warming, which varies largely between short- and long-lived pollutants. The recently proposed GWP* copes with this gap, accurately indicating the emissions' impact of GHG. Using GWP100 and GWP*, we calculate and compare the effect of CH₄ emissions from 2010 to 2020 of the Italian livestock production, starting from the official emission data published by Istituto Superiore per la Protezione e la Ricerca Ambientale (ISPRA) from 1990 to 2020. GWP100 and GWP* were assessed as follow: i) CO₂e = ECH₄(t)×28; ii) CO₂we = 28×[4.53×ECH₄(t)-4.25×ECH₄(t-20)] where ECH₄ is the annual CH₄ emission for a considered year(t), or relative to the previous 20 years (t-20). Except for buffalo (+ 240.7% of emissions calculated with GWP*), almost all species showed lower GWP* than GWP100, with the greatest re-dimensioning for non-dairy cattle (-47,263 kt of CO₂we and +66,437 kt of CO₂e, GWP* and GWP100, respectively). A huge difference was observed also for the total cumulative contribution of Italian livestock production to global warming over the past 10 years, including the nitrous oxide (N₂O) emissions, with GWP* and GWP100 accounting for -41,057 kt of CO₂we and +206,091 kt of CO₂e, respectively. In conclusion, the application of GWP* metric to CH₄ emissions of all Italian livestock supply chains allows to better identify the role of Italian livestock on climate change, and to better assess the contribution of this sector to offset global warming, thanks to the improvement of the production efficiency.

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Development of a Simplified Tool for Assessing Climate Change Impact in Dairy Cattle Farms

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The international literature strongly advocates for the adoption of the Life Cycle Assessment (LCA) method for evaluating environmental impacts, despite its high cost due to extensive data requirements. This study aims to develop a simplified tool for estimating climate change (CC_{es}) of milk production. The tool is based on 54 dairy farms in Northern Italy, where LCA analyses were performed, and fertility and efficiency data were collected. The CC_{es} was derived through a linear model and stepwise selection (Ordinary Least Squares, OLS), with a P-value for variable inclusion/exclusion set at 0.2. Variables with a high Variance Inflation Factor (VIF) were excluded. Selected parameters were biogas, soybean in the diet, Economic Health and CH₄ index, age at first calving, pregnant cows at 120 d, and milk sold per Livestock Unit. The adjusted R² of the equation was 0.63. Validation required the random selection of 15 farms from the database, repeated 1,000 times, resulting in an average correlation coefficient between CC_{es} and observed CC of 0.77. The average observed CC was 2.00±0.31, while CC_{es} was 1.93±0.26 kg CO₂ eq/kg FPCM.

Development of Mitigation Engine for specialized dairy farms

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Climate change is one of the main challenges the world has ever faced, and livestock production plays a role in it by emitting greenhouse gases (GHG). Efforts have been initiated to reduce the GHG of dairy systems. To achieve this, four main steps are essential namely, i) monitoring GHG emissions using available tools, ii) comparing the farm-specific emissions with the company or national targets, iii) defining measures using decision support tools, iv) implementing mitigation measures. At the steps of selection and implementation of mitigation measures, farmers need various supports. Dairy production is a complex system and any change in it results in lots of consequences. There are interactions not only between mitigation measures but also between measures and production factors. This requires detailed modelling at the farm scale. The Mitigation Engine (ME) is constructed as a meta-model, not simulating farm situations completely, but based on farm models and literature, calculating the interactions and trade-offs. ME is based on the principles of life cycle assessment (considering both on- and off-farm emissions) and using IPCC and NEMA calculation rules. ME is constructed to load output from monitoring tools to develop tailored recommendations. The GHG reduction potential and economic impacts of twenty mitigation measures have been modelled in ME. The first version of ME has been developed in an Excel environment. Using the farm-specific data imported automatically from the Dutch national monitoring tool (ANCA), various mitigation measures can be selected and by identifying the level of changes, ME quantifies the GHG reduction potential. ME is being tested in a couple of research projects to quantify the GHG reduction potential of dairy farms in the Netherlands. The case results of testing ME in a farm with 207 dairy cows with an average annual milk production of 9,008 kg/cow showed that the implementation of eight measures can reduce the carbon footprint of a dairy farm from 1,146 g CO₂eq/kg FPCM to 739 g CO₂eq/kg FPCM which is equivalent to 35% reduction of farm emissions. Some of the advantages of ME include: i) consideration of the interactions of measures, ii) user-friendliness, iii) integration with a monitoring tool, iv) farm-specific GHG reduction potential.

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Inclusion of a robust estimation method of carbon soil sequestration in the carbon footprint analysis of dairy sheep farms

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The aim of this study was to estimate the carbon footprint (CFP) of dairy sheep farms including in the CFP analysis a robust method to estimate soil carbon (C) sequestration. Six medium (M; 500 head, 50 ha) and six large (L; 100 ha, 1000 head) size dairy sheep farms located in Sardinia region (Italy) were considered. Data were collected by interviewing the farmers, in a cradle-to-farm gate approach based on ISO14040:2006 and 14044:2006. The time boundary selected was a period of 12 months (from October 1st to September 30th). Emissions were quantified according to Tier 2 of the IPCC (2019), while C sequestration was estimated according to the Petersen et al. (2013) method. Greenhouse gas emissions (GHG) were expressed in CO₂ equivalent units assuming a 100-year time horizon (27.9 for CH₄, 273 for N₂O, and 1 for CO₂). The considered functional units (FUs) differed in relation to the main product (milk: one kg of fat and protein corrected milk, FPCM) and co-products (meat: one kg of live weight sold, LWS; wool: one kg of greasy wool, GW). Without the inclusion of C sequestration, the CFP of milk was 2.52 and 2.76 kg CO₂e/kg FPCM, for M and L farms respectively. Concerning the co-products, the CFP was: a) 8.07 and 7.81 kg CO₂e/kg LWS, for suckling lambs in M and L farms; b) 0.46 and 0.44 kg CO₂e/kg GW, for wool in M and L farms; c) 1.22 and 1.36 kg CO₂e/kg LWS, for culled sheep in M and L farms. When soil C sequestration was included in the model, only the emissions associated with meat and greasy wool were completely offset, reaching carbon neutrality. The inclusion of an indirect robust method to quantify soil C sequestration can improve the CFP estimation and could be a valuable alternative to expensive and laborious direct methods of C soil quantification.

A preliminary overview of methane emissions in the Italian dairy sector using DHI data

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Methane is a transient gas that lasts for about a decade in the atmosphere, contributing approximately 0.5°C to observed global warming. Particularly, the dairy sector accounts for approximately 30% of the GHG emissions of the livestock sector and 4% of the total anthropogenic emissions. Approximately 80% of emissions originate from enteric methane. Enteric methane emissions were estimated by analyzing Dairy Herd Improvement (DHI) in 8368 Holstein dairy farms in Italy. The overall enteric methane emission averaged 0.023 kg CH₄ per kg fat and protein-corrected milk production (FPCM) yield and ranged from 0.014 to 0.099. We detect a negative correlation between CH₄ emissions and herd size and milk yield using kg FPCM as a functional unit and a positive CH₄ correlation expressing emissions as kg CH₄ per Livestock Unit per day.

Circles EU project: Slurry treatment with a protocol of bioactivators can reduce air and wastewater pollution in growing pigs

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Mitigating the ammonia (NH₃) emissions from manure is crucial for pig production, as high levels of NH₃ can have a negative impact on the environment, pigs' health and welfare. This study investigated the impact of the application of bacterial-enzymatic bioactivators to reduce on-farm greenhouse gas emissions, wastewater characteristics, and pigs' welfare during the growing phase. 980 pigs (average BW of 47.14±3.8 kg), were randomly assigned into a control group (normal farming) or a treated group (TR), in which 3 interventions were used: 1. Tablet supplied through the slatted floor into the wastewater tank (1 tablet/week, every 2 m³ of the slurry pit); 2. Powder sprinkled on the empty tank floor (1 Kg/week/10.000 kg of animal weight); 3. Liquid dispersed via an automatic sprinkler system. The groups were housed in a separate room (2 pens/room/group) equipped with a slatted floor, mechanical ventilation, and a pen wastewater tank. The trial lasted 63 days. Wastewater samples were collected before each draining (d21, d42, d63) for quality and microbiota analysis and air quality, and lesions on the ears and tails of pigs were recorded. A factorial design including the effect of treatment, sampling time, and their interaction as factors was applied. The wastewater of the TR group had lower pH (P=0.03), total (P=0.02) and organic (P<0.001) N, NH₄ (P=0.04) and a higher total K₂O (P<0.001). The air in the TR group had a lower NH₃ (P< 0.001). There was no effect on ear and tail lesion scores. In conclusion, the on-farm application of bioactivators increases the wastewater quality and mitigates NH₃ emissions of pig farms.

Methane emissions of two contrasting pasture-based dairy systems in Uruguay

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This study evaluates Methane (CH₄) emissions from dairy cows (Holstein and Holstein × Jersey; 465 Kg PV ± 59) grazing two systems with differing intensification levels. The trial was conducted at the Southern Regional Centre of the Faculty of Agronomy (UDELAR, Uruguay) during winter (P1) and spring (P2) 2023. The systems studied were BC, with a stocking rate (SR) of 1.8 LU/ha, and self-sustained, prioritizing operational simplicity; and AP, with an SR of 3 LU/ha, focusing on maximising productivity. The CH₄ emissions were determined by the SF₆ tracer technique, alongside milk yield (MY), milk composition, and pasture quality assessments. Fat and protein-corrected milk (FPCM) was estimated as proposed by IDF (2019). Individual DMI was estimated based on the model by de Souza et al. (2019). A linear mixed model in R was used for statistical analysis. Preliminary findings suggest a tendency towards higher CH₄ emissions in AP in both periods (P = 0.08; 292 and 278 g/day for AP and BC respectively). However, no significant differences between systems were observed in other variables such as DMI (P = 0.94, 18.8 kg DMI/day), or CH₄ intensity (P = 0.38; 11 g CH₄/kg FPCM). Accurate emission factors tailored to specific systems are essential for reliable sustainability assessments. Although CH₄ emissions did not significantly differ, both systems show high efficiency with low methane intensity. Further study is underway to determine whether more intensified systems, such as AP would lead to an increase in the Carbon Footprint per unit area.

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Poster 14

Exploring Greenhouse Gas Emissions in Sub-Saharan Africa: Are Camels a Neglected Source?

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Camels are crucial to food security, livelihoods, economy and culture in many arid regions in Africa and they are increasingly being considered as an important species for adaptation to climate change due to their resilience to heat and drought. Given their low share in total livestock numbers, their greenhouse gas emissions are not typically assessed and only a few Tier 1 estimates exist. Here, we present the first Tier 2 assessment of direct emissions of the pastoral camel population in sub-Saharan Africa in 2021 (33 million heads) and related to the production of protein from camel milk and meat. We used animal numbers from FAOSTAT and the DYNMOD herd model to estimate the herd structure, calculating cohort-specific energy requirements, and feed intake according to the IPCC 2019 guidelines and GLEAM methodology. Feed quality parameters were retrieved from the literature on pasture composition. The weighted emission factor for enteric fermentation per animal (61, range: 31-83 kg CH₄/head/year) was slightly higher than the value recommended by IPCC for the Tier 1 approach (46 kg CH₄/head/year), highlighting the need to use Tier 2 approaches whenever appropriate data are available. Estimated total GHG emissions for the sub-Saharan Africa camel population amounted to 56.2 MtCO₂eq/year, with 99% stemming from enteric fermentation and 1% from manure deposition on pastures. These represent a significant share of all direct livestock emissions in sub-Saharan Africa, suggesting the need to further investigate and integrate these species in national GHG inventories and Nationally Determined Contributions.

Analyzing Seasonal Emissions from Goat and Cow Manure in Cyprus

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Abstract The surge in global meat and dairy consumption has increased farming, consequently escalating manure production. Manure is a critical source of ammonia (NH₃) and greenhouse gas (GHG) emissions which are critical contributors to global warming. This study provides a comprehensive assessment of these emissions from dairy cow and goat manure in Cyprus during the summer season, aiming to elucidate the emissions dynamics and their driving factors. Dried manure samples collected from the Agricultural Research Institute farm in Athalassa (Nicosia, Cyprus), were added into ten polystyrene boxes, five for each animal species in a completely randomized design. GHG and NH₃ emissions were measured with automated non-steady-state (a-NSS) chambers and were analyzed with a cavity ring-down spectrometer (CRDS). Fifteen days after the initiation of the experiment, a simulated summer rainfall event was introduced to assess the impact of moisture on emission rates. Manure samples were taken before the initiation of the experiment, before the simulated raining event (T₂), a day after the event (T₃) and at the end of the experiment. At each sampling point, physicochemical properties and functional bacterial guilds involved in GHG were assessed. Our findings revealed that the GHG emission pattern was different between cows and goats, likely due to differences in the composition and digestibility of the feedstock used. Before the simulated rainfall, emissions were low in the dry manure conditions typical of Cyprus's summer. The addition of water increased NH₃, CO₂, and N₂O emissions, while CH₄ emissions decreased sharply. The abundance of key bacterial communities involved in CH₄ and N₂O emissions changed over time and the increase of water content had a substantial effect on these guilds. This study highlights the pivotal role of moisture in regulating GHG and NH₃ emissions from livestock manure, particularly under extreme temperature conditions. These insights are crucial for developing strategies to mitigate emissions in the context of global climate change.

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Theatre 1

Decision Support System for Insects Production

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Insect value chains are complex systems with non-linear connections among various economic, environmental, and social variables. Industrial stakeholders often require clarity in potential environmental, social and economic issues for efficient decision-making. While multiple studies have explored individual aspects of sustainable insect chains, like rearing or post-processing, there is a lack of how different industrial choices affect these chains, leading to a gap in their integration into industrial practices. This study employed a modular life cycle approach to analyze insect production chains, considering economic, environmental, and certain social aspects within the frameworks of the EU projects SUSINCHAIN, ADVAGROMED, CIPROMED and GiantLeaps. Integration of results from separate modules was based on developed scaling factors, further used to design multiple production scenarios, employed for multi-objective optimization (MOO) and the design of a decision-support system (DSS). The online open access DSS that based on the MOO with key defined objectives allowed us to define a limited number of optimal scenarios, reflecting on the optimal scenarios for location, insect species, size of industries and feed mix. Moreover, MOO allows for identifying a larger range of optimal scenarios with varying objectives. Further research aims for a broader spectrum of optimal scenarios, alternative protein sources and their validation.

Safe insect rearing on residual streams

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The four-year project SAFE INSECTS has studied the safety and feasibility of rearing black soldier fly larvae (*Hermetia illucens*) and yellow mealworms (YMW, *Tenebrio molitor*) on several residual streams. The residual streams included were supermarket mix, poultry manure, slaughter by-products and vegetables, fruit and food waste from households. Insects can convert these low quality streams into protein-rich ingredients for food and feed. However, chemical hazards, pathogens and viruses could be present in these residual streams. Under current European law, it is not permitted to use residual streams that contain animal products or manure for insect rearing. Therefore, necessary safety data is needed to provide a basis for amendment of legalization. Chemical and microbiological safety parameters were included in the experiments such as heavy metals, pesticides, dioxins, microplastics, food pathogens, parasites, viruses, and antimicrobial resistant genes. Experiments were performed with natural contaminated residuals streams and spike experiments were performed with pathogens, viruses and AMR genes. The main results related to food and feed safety will be presented. In conclusion, the food safety issues were different for different residual streams used as insect feed, and mitigation opportunities were studied. With our results we aim to deliver input to (re)evaluation of the possible use of these residual streams as insect feed.

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Theatre 3

Determination of microplastics in reared black soldier fly larvae (*Hermetia illucens*) and yellow mealworms (*Tenebrio molitor*) using polarized light optical microscopy

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Insects are increasingly seen as an alternative source of protein for both animal feed and human consumption, while also showing potential for sustainable and circular bioconversion of residual streams. Former foodstuffs contaminated with packaging materials could provide an alternative type of feed material for insects. These are not suitable for conventional livestock animals, due to animal welfare concerns (choking, accumulation) and potential exposure for the final human consumers. We hypothesize that such concerns may not, or to reduced extent, be applicable to reared insects. Lack of data has restricted a risk assessment and additional research is needed to fill current data gaps. In two different experiments, black soldier fly larvae (BSFL, *Hermetia illucens*) and yellow mealworms (YMW, *Tenebrio molitor*) were reared on substrates spiked with different types of plastics representing packaging materials to determine the presence of microplastic materials in the larvae. Larvae were processed (digested and filtered) to enable detection of microplastics, using polarized light optical microscopy. PET and LDPE were included in both experiments, while more polymer types were added for YMW. LDPE particles appeared virtually absent in BSFL, while PET had been transferred to the larvae. For YMW, more LDPE particles were found than PET and several aspects appear to have an influence on the transfer of additionally tested particles. Results from these experiments serve as the starting point for a new research project (2024-2028) to gather more data to obtain more insight on the rearing of insects on feed materials containing packaging materials, including biodegradable packaging materials.

Chemical and microbiological safety of black soldier fly larvae reared on sewage and industrial sludges

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The utilisation of black soldier fly larvae (BSFL, *Hermetia illucens*) for the valorisation of diverse residual streams is receiving increasing attention. In general, BSFL are efficient converters of waste into protein-rich biomass, with the resulting frass used as fertilizer. Sludges from domestic wastewater treatment plants and food processing industries have the potential to be integrated into the circular food system to recycle nutrients within the food system. These sludges might also be suitable as substrates for BSFL rearing. However, the potential use of these sludges for BSFL rearing requires attention to the impact on food safety due to the possible presence of various hazards. This study aimed to gain insight into the fate of selected chemical and microbial hazards within domestic sewage sludges and industrial sludges used for BSFL rearing, and their subsequent presence in BSFL and frass. Analyses of heavy metals and dioxins were thus far included, as well as microbial hazards (bacterial pathogens and antibiotic resistance by ESBL- and carbapenemase producing bacteria). The results of these safety analyses will be presented. So far, varying amounts of chemical and microbial hazards have been detected in the examined sludges, BSFL and frass. Moreover, not all studied microbial pathogens have been detected in the sludges nor appeared to transfer to BSFL or the frass. Overall, the results of this study provide knowledge on the potential impact and application of sewage and industrial sludges in BSFL rearing, particularly on their safety.

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Theatre 5

Investigating an invisible enemy: the problem of ammonia in insect farming

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Insect farming is blooming because these organisms can provide excellent nutrients at low environmental cost. Beyond requiring less water, land, and feed per kilogram of protein produced, insects produce also considerably less greenhouse gases than traditional livestock. Ammonia (NH₃) is a gas that can result from insect farming activities. Type and pH of substrate used to rear the larvae can influence the release of this gas in the air since the dissociation of the ammonium ion (NH₄⁺) occurs at high pH. Although ammonia emissions of insect farming can be lower than traditional livestock, they can still be detrimental for the environment, insect breeders, but also insect survival. Nevertheless, there is no evidence nor quantification of potential lethal effects on larvae induced by direct contact or exposure to these chemicals. Little has been explored with respect to the quantity and potential effects of ammonia emissions linked to substrate formulations. Therefore, there is an urge to: 1) Develop methods to quantify the NH₄⁺ and NH₃ content, particularly in the substrate, 2) Identify potential origins of NH₃ emissions, 3) Understand the (sub) lethal effects of short and long-term exposure to both NH₄⁺ and NH₃ on larvae, and 4) Identify and implement prevention actions to counteract the production of ammonia in insect mass rearing. We began to investigate these aspects in the black soldier fly (BSF) with particular attention to the effect of 13% concentrated commercial ammonia both in gaseous form (NH₃) or diluted in the substrate (NH₄⁺; from 1:1 to 1:6 ammonia:water v:v). Preliminary results show that BSF larvae are adversely affected by both forms. All larvae died after three hours of concentrated NH₃ exposure, while 1:1 and 1:2 NH₄⁺ concentrations reduced survival (52.94 and 73.53% respectively) with respect to average (92.40%). More research is needed on this topic particularly considering its effects on insect survival, breeders, as well as the environment. This information is critical to anticipate regulatory aspects and design insect mass rearing that limit ammonia emissions.

Sustainability of complex insect production chains: from food waste to feed and food

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Insect production for food and feed is facing numerous challenges associated with potentially high costs, environmental impact and a lack of optimization in production. Particularly, these limitations are associated with the legal limitation in EU on the feeds allowed for insects as farmed animals. Considering the challenges, the need in the holistic assessment approach (Life Cycle Assessment) of suitable agri-food side streams is needed. LCA was cradle-to-grave analysis with IMPACT 2002+ method built on systemised inventory of insect production systems in the frame of Advagromed and Cipromed projects. Literature on LCA of organic wastes or side streams used for insect production was also considered. The results indicated that while it is environmentally feasible to substitute conventionally produced fertilisers with insect frass, protein and lipid sources for feed and food with insect biomass, these substitution routes have different levels of circularity efficiency. Biotransformation of food waste with insects has a very low direct impact (around 1% of to the impact of the whole production chain). It was identified that insect chains can reduce environmental impact of agri-food chains by 4-15% and improve the nutrient recycling potential by 10-15% (nutrients returned into soils), by 30-38% for the nutrients returned in form of food products. The efficiency depends on the initial nutrient richness of the food waste, the biotransformation abilities of insects, place of nutrient return in the chain and on the upstream-to-downstream transformation of the nutrients along the production chain.

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Response to mechanical stress in yellow mealworms (*Tenebrio molitor*) with regards to welfare considerations

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Insect farming for food and feed has garnered significant attention as a fast-growing novel industry with the potential to address global food security challenges. Among the topics raised by this new sector, the welfare of farmed insects is now a factor that has started to be considered. This study investigates for the first time the impact of mechanical stress on *Tenebrio molitor*, as possibly experienced during on-farm rearing operations. A particular focus was given to stress generated by pressure in order to assess the critical levels that result in changes in : behavior, physical integrity, development, growth and reproduction, observed at different insect life stages (larval, pupal, and adult stages). First results show greater resistance to pressure for larvae and thereafter for adults, compared to pupae, with threshold of 500g, 250g and 50g respectively, indicating the maximal pressures required to avoid the risk of injury or mortality. Whatever the force applied, some behavioral changes were observed such as escaping or body twisting. However, no lagged effects were observed and behavior quickly returned to normal with no long-term impacts on metamorphosis, emergence or egg-laying rates, even for insects unharmed from high pressure. These first findings provide a novel and unexplored insight into the sensitivity of *Tenebrio molitor* to mechanical stress, thus allowing engineers to improve the design of rearing equipment while respecting the welfare of this farmed insect.

Enhancing Poultry Sustainability: Black Soldier Fly Effects on Laying Hen Welfare and Microbiota

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Historically chickens eat live insects while today poultry farming is based on soybean, linked to deforestation and GHGs emissions. The insect “Black soldier fly” (BSF) is emerging as alternative and sustainable protein source. We evaluated the effects of live BSF larvae supplementation on Lohman hens, in terms of feathering score (FS), microbiota (M), excreta corticosterone (EC). 108 birds (16-34 weeks (w) age) were housed in 27 cages and 3 groups: C (control) fed commercial feed, G1 and G2 fed commercial feed plus live BSF (15%-30% of the expected DFI, respectively). After monthly evaluations, FS, EC data were elaborated by “time”, “diet”, their interaction; while M informed on Alpha-Beta diversity, differential abundance. FS wasn't affected by diet ($P < 0.001$), EC concentration was influenced by diet and time being lower in G1-G2 than C ($P < 0.001$) and decreasing at the end of the trial ($P < 0.01$). Occurred higher Alpha-Beta M diversity in G1 and G2, with Lactobacillus and Clostridiales concentration positively affected by diet and time. The supplementation of BSF live larvae represents a sustainable protein source for the poultry sector, able to improve animal welfare.

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Welfare assessment of managed honey bee colonies: a preliminary study

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Animal welfare (AW) assessment is still a topic of great discussion as there is no consensus on the protocols used for livestock species and it is even more difficult to assess AW in farmed insects. This study aimed to develop an AW assessment protocol for honey bee (*Apis mellifera*) colonies to be included in the ClassyFarm IT platform of the Italian Ministry of Health. In Italy, ClassyFarm AW assessment protocols are currently used for official-controls and self-controls in the main livestock species. In ClassyFarm, the on-farm AW measurement is carried out by means of a multifactorial approach including both resource-based (RBIs) and animal-based indicators (ABIs). For the purpose of this study, 16 Italian beekeeping experts were involved into a focus group to discuss a list of RBIs and ABIs, previously selected by the authors through a review of the scientific literature, in order to identify potential valid, feasible and reliable RBIs and ABIs for apiary welfare assessment. Despite the difficulties encountered related to the nature of honey bees and the limited availability of scientific knowledge about their welfare, 14 RBIs were selected: beekeeper training, number of hive inspections, colony equalisation, queen replacement, queen selection, artificial swarming, feeding, management of hives affected by diseases, anti-varroa treatments, overwintering, transport, culling of the colony, hive type and management, hive position and orientation. The experts also identified 7 ABIs: winter mortality, abnormal colony depopulation/mortality, robbing signs, number of drone laying queen colonies, number of queenless colonies, irregular brood pattern, clinical signs of varroosis. These indicators are currently tested on-field in Italian apiaries in order to gather data and formulate the final AW assessment protocol. The study was supported by the Italian Ministry of Health PRC IZSLER 2020/06 and PRC IZSLT 2022/08

Scaling Up Bioremediation of Livestock Manure Using Black Soldier Fly Larvae (*Hermetia illucens*)C. Ligeiro^{1,2}, A. M. Machado¹, R. Fantatto^{1,2}, L. G. Guilgur^{1,2}, D. Murta^{1,2}¹ *Ingredient Odyssey SA - EntoGreen, R&D, Rua Comendador José Júlio Eloy, 2005-332 Santarém, Portugal,*² *Egas Moniz Center for Interdisciplinary Research (CiiEM), R&D, Egas Moniz School of Health & Science, 2829-511 Almada, Portugal*

The global demand for food, has led to significant challenges in managing the livestock manure generated, posing environmental and public health risks. The exploration of innovative approaches like bioremediation using black soldier fly (*Hermetia illucens*, BSF) is needed. This study investigates the potential of BSF in bioremediating poultry manure through a large-scale trial. Building upon insights from a preliminary trial, the study aims to validate and expand upon initial findings by scaling up the process. By utilizing 144 crates per group, the experiment seeks to assess the feasibility of converting manure into valuable biomass. Key advantages of BSF larvae bioremediation include reduced greenhouse gas emissions, pathogen suppression and minimal water requirements. The study also evaluates larval weight, odour and substrate temperature, as well as microbiological and heavy metal assessments. Preliminary results indicate comparable trends in temperature evolution and larval biomass between control and test groups. The large-scale trial is critical for substantiating the efficacy of BSF larvae in poultry manure bioremediation. Its finding promises to revolutionize waste management practices, informing sustainable agricultural strategies that mitigate environmental risks and enhance productivity. This study was conducted under the scope of the FCT PhD scholarship UI/BD/154566/2023, and the projects Safe Insects LWV20.102 BO-64-001-025 and InsectERA – A ERA da indústria dos insectos C632327738-00466948, funded by PRR – Plano de Recuperação e Resiliência, RE-C05 – Agendas Mobilizadoras para a Inovação Empresarial, República Portuguesa.

Session 82

Theatre 1

By-products for small ruminants: benefits and limitations

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The great availability of agro-industrial byproducts produced at the local level, appears to be a promising strategy for reducing competition with human edible foods, the cost of out-farm produced feeds, and the environmental concerns of livestock production. The presence of bioactive compounds, such as polyphenols, confers added benefits to these byproducts when they are included in the diets of small dairy ruminants. In this presentation, we analyze the most recent scientific results concerning the use of agro-industrial by-products, naturally rich in polyphenols, in the diets of small dairy ruminants. Effects on colostrum and milk production, milk and rumen liquor fatty acid profile, metabolic parameters, and methane production are discussed. Different positive effects of byproducts have been reported on nutritional quality of milk, antioxidant activity in milk and blood, and rumen fermentation. The expected beneficial effects of dietary polyphenols in small ruminants were not always observed probably because of limitations due to large variability in chemical composition, nutrient content, and storage issues. However, owing to the large quantities of local products available at low prices, the use in sheep and goats nutrition offers a convenient solution to the valorization of residues arising from agricultural activities, reducing feed costs for farmers, and conferring added value to dairy products, in a sustainable way. To overcome these limitations the regular and consistent characterization of byproducts is crucial before their inclusion in a dietary formulation. NextGenerationEU; PNRR M4C2, CN00000022 AGRITECH.

Dehydrated sulla forage to improve sustainability of sheep milk production

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Grazing sulla forage, containing moderate levels of condensed tannins, is known to improve small ruminants' physiological status and productive responses. Preliminary investigations showed analogous positive effects with pelleted and dehydrated sulla forage (DSF) in the diet of dairy ewes. In this research, the DSF was compared with sulla hay (SH) in terms of milk production, feed utilization efficiency, and ewes' oxidative status. Using a 3×3 Latin square design, 9 ewes of Valle del Belice breed at 60 d in milk were fed a diet based on SH ad libitum (crude protein (CP) 8.8 % dry matter (DM)) or diets with 2 kg/d of DSF obtained by cuts in April (ADSF; CP 15.1 % DM) or May (MDSF; CP 11.3 % DM). Each ewe received 800 g/d of concentrate feed. Data were analyzed statistically using a MIXED model. The results showed how groups fed DSF increased DM intake (3.6, 3.6, 3.0 kg DM/d with ADSF, MDSF and SH; P<0.001) and milk yield (1.7, 1.6, 1.3 kg/d with ADSF, MDSF and SH; P<0.001), without reducing casein. The ADSF diet also improved efficiency in milk conversion and DM digestibility (72, 58, 50 % with ADSF, MDSF and SH, P<0.001). Due to higher intake, estimated daily methane emissions increased with DSF diets (45, 45, 38 g CH₄/d with ADSF, MDSF and SH; P<0.001), while was lower with ADSF if referred to milk yield (28, 30, 31 g CH₄/kg milk with ADSF, MDSF and SH; P<0.05). Moreover, feeding ADSF induced a higher plasma polyphenols level (27, 17, 19 µl GAE/ml with ADSF, MDSF and SH; P<0.05) enhancing the ewes' oxidative status. These results showed how dehydration of sulla forage cut in an early stage can represent a valid alternative to traditional hay in periods of low resources at pasture.

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Theatre 3

Carob germ: An alternative protein source in feeding lamb

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The increase in world population and eating habits require sufficient meat production, representing a serious challenge for the future. In this scenario, providing a huge quantity of imported feedstuffs needs to optimize production, although controversy regarding their sustainability. Therefore, the use of local by-products as alternative protein feed sources could be helpful to improve this aspect, reducing the environmental impact of livestock farming. To this purpose, this study aimed to assess the effects of including carob (*Ceratonia siliqua* L.) germ, cultivated in the Mediterranean area, in lambs' diet on animal performances and meat quality. Twenty-eight 40-day-old male lambs were selected from the native farm and randomly assigned to three groups. The 1st group was fed a conventional diet (CON), including maize (43%), hay (30%), soybean meal (22%), molasses (3%), and vitamin-mineral (2%). The 2nd and 3rd groups received the same diet but with 25% of carob germ as partial soybean meal and maize replacement (CAR25) or 50% as a total replacement of soybean meal and partial of maize (CAR50). All lambs were slaughtered in a local slaughterhouse after 56d of the trial. Carcasses were weighed, and stored at 4°C for 24h, after that Longissimus dorsi muscles were collected for subsequent analyses. Lambs' performance and carcass traits revealed no significant differences between the three experimental groups. Results on meat samples indicated that drip loss of CAR25 and CAR50 were significantly lower (P < 0.05) than CON after 2 and 4 days of storage, which may contribute to improve tenderness and juiciness. We hypothesized that dietary carob supplementation might decrease drip loss of lamb meat due to a possible increase in antioxidant activity. Further analyses are in progress to investigate this aspect. Acknowledgments: PRIN RE-F-RESH (CUP E53D2301105 0006).

Sodium bicarbonate limited milk fat depression of lactating dairy ewes fed ryegrass herbage rich in water soluble carbohydrates

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The use of grass pastures rich in water soluble carbohydrates (WSC) in ewes can induce ruminal sub-acidosis, resulting in a decrease in milk fat content. With the aim to prevent sub-acidosis conditions, the effect of supplying sodium bicarbonate, as rumen buffer, to sheep fed fresh cut ryegrass (*Lolium multiflorum* var. *westervoldicum*) herbage rich in WSC (mean of 27.7 % DM) was studied. Ten lactating ewes of the Sarda breed in their third month of lactation were selected, based on milk production and body weight. They were then divided into two homogeneous groups: control (CNT) and bicarbonate (BIC). The animals were fed indoors with automatic Biocontrol AS (Rakkestad, Norway) feeders, which recorded continuously during the day the individual intake of grass of the ewes. The ewes received ad libitum freshly cut grass from 1:00 PM to 7:00 AM of the subsequent day. In addition, they received, in two separate meals at milking (7:00 AM and 16:00 PM), a total of 400 g/d of SBM and 550 g/d (as fed values) of whole corn grains. The BIC group also received a supplement of 25 g/day/ewe of sodium bicarbonate just before the ryegrass supply. Data on intake and milk yield and composition were covariate-adjusted using pre-experimental measurements. Daily DM intake of grass (BIC 1488 g DM/d, CNT 1340 g DM/d; $P=0.03$), particularly high in the first hours after its supply, was higher in BIC than in CNT. Milk yield (CNT 1859 g/d, BIC 1838 g/d) was not affected by the treatment. Milk fat concentration of afternoon (BIC 6.52%, CNT 6.01%; $P < 0.001$) and morning (BIC 5.39%, CNT 5.06%; $P=0.03$) milking was higher in the BIC than CNT. Moreover, milk fat yield of afternoon milking was higher in the BIC than CNT (BIC 42.2 g/d, CNT 38.7 g/d; $P=0.03$) but there were no differences in morning milking (BIC 64.7 g/d, CNT 60.6 g/d). The fat to protein ratio of the afternoon (BIC 1.35, CNT 1.25; $P < 0.001$) and morning (BIC 1.13, CNT 1.06; $P=0.01$) milk was higher in BIC than in CNT. In conclusion, sodium bicarbonate supplied just before the grass supply showed a marked ability to reduce milk fat depression, especially in the afternoon milking, probably due to its stronger short-term action.

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Theatre 5

Total replacement of soybean meal with lupine seed in lamb diet: effects on growth performance

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Due to the growing consumption of meat, the demand for protein feed has increased in recent years. The use of local alternative protein feed sources may be a strategy to reduce environmental impact of feedstuff supply chain. In this study, we aimed at assessing the effects of inclusion of lupine seeds (*Lupinus albus* L.), cultivated in Mediterranean region, in lambs' diet on animal performances. Sixteen male lambs (40 days old; 12.8 ± 2.33 kg live weight) were selected from the native farm and randomly allocated to two experimental groups. A 1st group was fed a conventional maize-based diet (CON), which included: maize (43%), hay (30%), soybean meal (22%), molasses (3%), and vitamin-mineral (2%). The 2nd group received the same diet but with 35% of lupine (LUP) as total replacement of soybean meal and partial of maize. On day 56 on feeding trial, lambs were weighted and sacrificed in local abattoir. Carcasses were weighted 24 h after slaughter and Longissimus dorsi muscles were collected for subsequent analyses. Daily intake of LUP diet was equal to 36% of that of CON one ($P < 0.001$). At the end of the feeding trial, the body weight resulted significantly lower in lambs fed LUP than CON diet (14.5 vs 27.1 kg respectively; $P < 0.001$). Indeed, during the trial the weight gain was only 2.1 kg in LUP lambs vs 13.9 kg in CON ($P < 0.001$). Consequently, carcass of LUP lambs weighed approximately half of CON lambs (11.9 vs 6.05 kg respectively; $P < 0.001$). We hypothesized that alkaloids present in lupine seeds had reduced palatability of LUP diet. Further analyses are ongoing to clarify this hypothesis. Acknowledgement: Agritech National Research Center.

Effect of feeding prickly pear by-products silage on dairy ewe's performance

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A total of 12 ewes (60 days in milk, DIM) were selected, and randomly divided into three experimental groups homogeneous for parity (> third-lambing), live weight (53.66 ± 6.57 kg), and milk yield (1.038 ± 144 g/day). Each group was fed for 14 days (9 for diet adaptation + 5 for sampling) with one of three experimental diets (3 groups \times 3 diets) according to a Latin square design. The diets were formulated to be similar for energy and protein contents (CTR: 900 g/d/head of commercial concentrate + Sulla hay ad libitum; PPP: 1.0 kg/d/head of silage (prickly pears peels + 12% of wheat bran) + 500 g/d/head of commercial concentrate + Sulla hay ad libitum; PPS: 1.5 kg/d/head of silage (prickly pears peels, pulp, seeds + 12% of wheat bran) + 500 g/d/head of commercial concentrate + Sulla hay ad libitum. The results showed that daily milk yield ($p < 0.05$) was lower when ewes fed PPP and PPS diet (658, 673 vs. 774 g/d with PPP, PPS vs. CTR). Nevertheless, milk protein ($p < 0.07$) and casein contents ($p < 0.05$) were higher with PPP than CTR diet (crude protein 6.29 vs. 5.86 %; casein 4.89 vs. 4.48 %, for PPP and CTR). The administration of both diets with prickly pears silages resulted in lower milk urea levels compared to the control group (38 and 40 vs. 49 mg/dl). Moreover, dry matter intake (DMI) was significantly lower when ewes fed PPP diet ($p < 0.01$). It was concluded the addition of prickly pears by-products silages in the diet of dairy ewes could be used to reduce the feed cost.

Sheep grazing preferences on pastures in the Montado: effect of soil amendment and grazing systems

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The aim this study was to evaluate the effect of dolomitic limestone application (uncorrected, U and corrected, C) and two sheep grazing systems (continuous, CG and deferred, DG) in preferred grazing areas, in natural pastures. The stocking rate was of 7 sheep/ha in CG and 16 sheep/ha in DG. In 4 plots of 1ha each, four treatments were constituted: T1 (UCG); T2 (UDG); T3 (CDG); and T4 (CCG). The study was carried out in Mitra farm at the University of Évora, between March and May of 2021. In beginning of March, mid-April, and end of May the animals were observed on the field and their location was recorded. The observation was carried out, in two consecutive days, by trained observers equipped with binoculars, from sunrise to sunset, every 10 minutes. In each plot, 12 georeferenced areas were defined, representing different pasture botanical communities. The day before observation, pasture height was measured. The results showed similar grazing patterns between treatments, with preference for lower altitude areas. Throughout the pasture's vegetative cycle, the grazed areas were expanded to higher altitude areas (April), then returning to lower altitude areas at the end of May. The height of the pasture was not a limiting factor that led animals to expand their grazing areas. Pasture botanical species did not prove to be decisive in the sheep's grazing choices. The results suggest that the application of dolomitic limestone and the grazing systems tested did not influence the sheep grazing preferences when pasture growth rate is maximum.

Composition and fatty acids profile of colostrum, transition milk and mature milk in dairy ewes fed grape byproduct during late gestation

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The objective of this study was to characterize the main composition and the fatty acid (FA) profile of colostrum (M1), transition milk (M2), and mature milk (M3) of sheep fed a small amount of grape pomace (GP) during the late gestation. The trial was conducted using 22 ewes in late gestation (15 days \pm of lambing) divided in two groups: CON, without GP supplementation, and GP with an inclusion of 50 g/d of GP. Animals were kept all together in a barn equipped with 10 individual automatic feeding system (Biocontrol AS, Rakkestad, Norway). M1 was collected within 12 h of lambing, M2 after 48 h, and M3 after 7 days. Data were analyzed using mixed model in which the diet, the sampling, and their interactions were used as fixed effects and the animals as random. The diet did not affect the main composition and the FA profile of the M1, M2 and M3. On the contrary, the sampling influenced almost all milk parameters, except the log of SCC. Fat and protein concentration was higher in M1 (10.42 and 14.63%, respectively) than M3 (5.43 and 5.42 %, respectively), whereas the lowest lactose concentration was found in M1 (3.03%). M1 had the highest NaCl concentration, due to the physiological animals' conditions after the lambing. As regard the FA profile, short chain-FA were lower in M1 compared to M2 and M3 (5.71, 7.94, and 13.46 g/100 g of FA). In contrast, the medium-chain-FA were higher in M1 than M2 and M3. In particular, C14:0 and C16:0 were 30% and 23% higher in M1 than M3, respectively ($P < 0.05$). Among long chain-FA, C18:0 was lower in M1 than M2 and M3 (-35% and -36% respectively), whereas C18:1 cis9 was higher in M1 and M2 than M3 ($P < 0.05$), evidencing a high fat mobilization that occurs during the first hours after the lambing. M3 had higher proportion of odd- and branched-chain FA than M1. In conclusion, colostrum, transition and mature milk are deeply different in composition and FA profile, reflecting the strong physiological change that occurs during and after the lambing. Acknowledgements: This work was financially supported by NextGenerationEU; PNRR M4C2, CN00000022 AGRITECH.

Inclusion of poplar leaves and stems in ewes' diet: effect on milk yield and quality and oxidative status

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The reduction of pasture biomass due to increasing extreme summer droughts and rising production costs negatively impact sheep milk production in the Mediterranean. However, using tree leaves and stems as fodder during dry seasons could be an interesting solution. This study aims to investigate the effect of including poplar leaves and stems into lactating ewes' diets on milk yield and quality. In June 2022 and 2023, an on-farm pilot trial was carried out by grouping six lactating ewes into control and treatment groups with three animals each. For 14 days the control group received a standard diet, while in the treatment group diet 0.5 kg DM of alfalfa hay was replaced with the same amount in terms of DM of fresh poplar leaves and stems daily harvested. Individual daily feed intake and milk production were measured, and milk samples analysed for fat, protein, urea, lactose, and fatty acids profile and antioxidant status. No significant differences were found in daily individual dry matter intake and milk yield and composition between treatments. Principal component analysis and stepwise discriminant analyses allowed to clearly separate milk from the two groups, being milk samples obtained from sheep fed with poplar leaves and stems higher in polyunsaturated fatty acids. The analysis performed on milk samples for the evaluation of the antioxidant status of the animals showed no difference in oxidative stress.

Effect of forage source (peanut vine and extruded rapeseed stalk) on growth, nutrient digestibility, rumen fermentation parameters of fattening lambs

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Rapeseed (*Brassica napus* L.) is the major oilseed crop in China and accounts for about 20% of world production. The proportion of rapeseed stalks used as ruminant feed is relatively low due to its poor palatability. Extrusion process could increase the palatability and digestibility of stalks with only slight increase in cost. In this study, we compared the effect of peanut vine (PV, a relatively high-quality forage) and extruded rapeseed stalk (ERS) on intake, apparent nutrient digestibility and ruminal fermentation parameters in fattening lambs, with an aim to provide insight into the utilization of rapeseed stalk as an economic forage source in ruminant production. Twenty-four weaned male Hu lambs (3-month-old) with similar body weight (20.0 kg) were fed diets containing either 70% concentrate + 30% PV or 70% concentrate + 30% of ERS. The diets were made isocaloric and isonitrogenous and the concentrate was composed of corn, soybean meal, wheat bran, salt, vitamins, and minerals. Each lamb was fed individually and had ad libitum access to diets and clean water. A t-test was used to determine if there was a significant difference ($P < 0.05$) in any variables. No difference was observed in dry matter intake (1.34 vs 1.35 g/d) and average daily gain (224 vs 212 g/d) of lambs fed either PV or ERS ($P > 0.05$). Apparent digestibility of nutrients was not different between two groups of lambs ($P > 0.05$). A higher ruminal total VFA production, molar proportion of propionate, but lower molar proportion of acetate and butyrate was identified in lambs fed PV compared with those fed ERS ($P > 0.05$). In conclusion, extruded rapeseed stalk as the only forage source did not negatively affect the growth performance.

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Poster 11

Carcass traits in lambs from local Shumen copper-red sheep, slaughtered at weaning in 90 days of age

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The Shumen copper-red sheep is a dual purpose native Bulgarian sheep breed. In the traditional rearing technology, the lambs has been sold for meat immediately after a suckling period (usually 3 months), and after weaning the ewes are milked for 2-3 months. In present work the carcass traits of 3 months old Shumen copper-red lambs has been investigated. The farm practices an extensive rearing technology, with almost year-round pasture, with additional feeding of the sheep only during the winter months. The newborn lambs are not separated from their mothers and during the suckling period the main food is mother's milk, and after the 10th day of birth, the lambs has free access to alfalfa hay, and concentrated feed (maize, barley). After 15 days of age, the lambs go out together with their mothers to the pasture, while the supplementary feeding continues throughout the all suckling period. A 6-th males lambs, born as singles, were slaughtered immediately at weaning, at 90 days of age, without a period of intensive fattening after weaning. Based on the results of slaughter analysis of suckling lambs from Shumen copper-red sheep, it can be concluded that the average slaughtered live weight was 22.9 kg, the average cold carcass weight was 11.9 kg. Dressing out of chilled carcass, without the offal was 51.9 %. The linear measurements of the carcass were as follows – diagonal carcass length – 52.3 cm, ham length – 32.0 cm, ham girth – 32.6 cm. The ratio meat:bones in the carcass of the Shumen lambs, slaughtered at weaning in 90 days of age, was relatively high – 3.15:1.

Effect of the inclusion of ensiled pomegranate by-product in the diet of dairy goats on milk production and its macro-composition

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The aim of this experiment was to study the effect of the inclusion of different proportions of the pomegranate by-product (*Punica granatum* L., var. Wonderful) silage in the diet of goats on individual daily milk production and its macro-composition. The effect of the inclusion of 0, 5, 10 and 15% DM in isoenergetic and isoproteic diets was studied. 80 goats were selected and distributed into 8 groups of 10 animals each (4 treatments x 2 plots) with homogeneous physiological characteristics. After the pre-experimental sampling, 4 more samplings were done fortnightly: daily milk production (Lactocorder®, WMB AG, Switzerland), milk composition (fat, crude protein, lactose, cheese extract, urea, MilkoScan FT6000, Foss Denmark) and somatic cell count (Fossomatic 5000, Foss, Denmark) were analyzed using a mixed linear model (Proc. Glimmix, SAS v9.4), considering the effect of the covariate in the pre-experimental sampling, the treatment (4 levels), experimental sampling (4 levels) and their interaction (16 levels) and the flock nested to the treatment, and the random effect of the goat. It was observed that only sampling ($P<0.0001$) and covariate ($P<0.0001$) had a significant effect. A decline in milk production was observed as the experiment progressed, while the variables fat, crude protein, cheese extract increased, and lactose and urea decreased. SCC decreased at the end of the experiment at the treatments with a higher inclusion (10 and 15%). It is concluded that pomegranate var. Wonderful ensiled can be included in the dairy goats diets up to 15% DM at short term, and long-term experiments are needed. This study was supported by Valencian Government. CIAICO/2021/301

Evaluation of energy content and protein value of tomato pomace for feeding ruminants

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Tomato pomace (TP) is a pollutant wet by-product of the tomato industry but can be used in ruminant feeding. This work aimed to evaluate the energy content and protein value of 12 samples of TP obtained from 2 Spanish processing plants at different sampling moments. Samples were dried, analyzed for crude protein (CP) content, and incubated in vitro with buffered ruminal fluid from 4 sheep for 24 h to measure gas production. Moreover, CP ruminal degradability (CPRD) was measured in situ after 16 h of incubation, and CP intestinal digestibility of the rumen undegraded CP fraction (CPid) was estimated in vitro following a three step-procedure involving treatment with pepsin (1 h) and pancreatin (24 h) solutions. Metabolizable energy (ME) content was estimated from chemical composition and gas production at 24 h, and protein value (PDIA, PDIN and PDIE) was calculated using INRA-2007 equations. The average ME of TP (10.3 ± 0.22 MJ/kg DM) was similar to that of INRA (10.1 MJ/kg DM). The mean CP content of TP was 160 ± 4.1 g/kg dry matter (DM; 131 – 190 g/kg DM), being lower than that provided by INRA (205 g CP/kg DM). Average CPRD and CPid values were $51.0\pm 1.82\%$ (41.0 – 60.9%) and $46.0\pm 0.84\%$ (40.1 – 49.9%), which were also lower than those provided by INRA (66 and 55% for CPRD and CPid, respectively). Mean values of PDIA, PDIN and PDIE were 39.9, 82.4 and 103 g/kg DM, respectively, whereas INRA reported averaged values of 43.0, 116 and 79 g/kg DM. Variations in CP content and the lower values of CPRD and CPid in our samples could possibly be due to differences in the industrial process generating TP.

Effects of feeding almond hulls to light lambs on meat quality and fatty acid profile

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Almond hulls (AH) are the most abundant by-product of almond production. Although AH are frequently fed to dairy cows, information on their use in lambs diet is limited and therefore the aim of this study was to analyze the effects of including increasing amounts of AH in the concentrate (0, 6 and 12%) of light lambs on carcass and meat quality parameters. Thirty Manchega lambs (15 males and 15 females) were distributed into 3 homogeneous groups according to their body weight (BW) and sex, and each group received one of the experimental concentrates and barley straw ad libitum. Lambs were slaughtered after 47 days of fattening at about 23 kg of BW. Including AH in the concentrate did not affect ($P \geq 0.175$) either BW at slaughter or hot and cold carcass weights. Meat pH and chemical composition were unaffected ($P \geq 0.052$) by the concentrate, but the meat of lambs fed AH6 and AH12 had greater proportions of polyunsaturated fatty acids (PUFA) than that of control lambs, indicating a healthier fatty acid profile for humans. The meat of AH12-fed lambs had greater concentrations of TBARS after 6 days of storage than that of AH6-fed lambs, possibly due to its higher PUFA content. In summary, AH can be included in the concentrate of fattening light lambs without negatively affecting growth performance. Almond hulls can improve meat lipid profile, but lipid oxidation can be increased.

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Poster 15

Influence of feeding agroindustrial by-products to sheep on ruminal fermentation and plasma metabolites

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The use of agroindustrial by-products in animal feeding could contribute to farm sustainability. The objective of this work was to study the effects of replacing conventional ingredients in a concentrate for dairy ruminants with a mixture of corn DDGS (dried distilled grains with solubles), dried citrus pulp (DCP) and exhausted olive cake (EOC) on ruminal fermentation and plasma metabolites in sheep. Two experimental diets based on alfalfa hay and concentrate (50:50) were fed to rumen-fistulated sheep in a cross-over design. A high-cereal control (CON) concentrate was formulated with corn, barley, wheat, soybean meal, wheat bran, palm meal and rapeseed meal (33.0, 20.0, 10.0, 12.2, 10.0, 8.8 and 2.5%, respectively), while in the BYP concentrate corn DDGS, DCP and EOC (18, 18 and 8%, respectively) replaced some feed ingredients in CON. Ruminal parameters were measured at 0, 3, and 6 h after feeding and plasma metabolites concentrations at 0 and 6 h. Sheep fed CON diet had higher ($P=0.012$) pH values than those fed BYP diet, but total volatile fatty acids (VFA) and $\text{NH}_3\text{-N}$ concentrations were unaffected by the diet. Feeding the BYP concentrate resulted in lower acetate proportions ($P=0.001$) and acetate/propionate ratios ($P=0.027$), as well as greater ($P=0.008$) butyrate proportions, compared with sheep fed the CON concentrate. No differences were detected between diets in plasma liver enzymes (GPT, GOT, GGT), while cholesterol levels were significantly increased in diets with BYP compared to animals fed the CON diet ($P<0.001$). The results indicate that these by-products can replace conventional ingredients without negatively affecting ruminal fermentation and plasma metabolites in sheep.

Evaluation of lactic acid bacteria as additive for wet brewer's grains and citrus pulp silage

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Several studies have reported a low ensilability of wet brewers' grains (BG), but BG could be mixed with other by-products of high ensilability. Lactic acid bacteria (LAB) are usually used as additive to increase silage quality for many crops. This study evaluated the effects of a LAB inoculant (*Lactobacillus buchneri*) on silage quality of a mixture of BG and dried citrus pulp (DCP). Wet BG was mixed with DCP in 3:1 proportion to reach 38% of dry matter (DM) content in the mixture. Twelve vacuum bags were filled with 500 g of the mixture: 6 bags were treated with LAB (1×10^6 CFU/g silage diluted in 100 ml water) and the other 6 bags (control) only received water. A vacuum-packing machine was used to extract the air inside the bags, which were heat-sealed and stored (dark; room temperature). After 3 months of storage, bags' content was analyzed for pH, DM and lactic acid concentration. Ensiling microbiology was assessed by classical plate-culture techniques. The addition of LAB did not affect pH ($P = 0.212$; 3.63 vs. 3.68), but decreased DM losses ($P = 0.029$) from 13.92% in control silages to 7.36%. However, LAB also decreased lactic acid concentrations from 49.15 to 29.39 g/Kg DM silage ($P < 0.001$). No *Clostridium*, *Escherichia coli*, *Listeria monocytogenes*, molds, *Salmonella* spp. and *Staphylococcus* coagulase-positive were detected in any silage. Whereas no *Enterobacteria* spp. were detected in LAB-silages, they were present in control silages (2.53 log CFU/g). Using LAB also reduced ($P < 0.001$) yeast counts from 5.67 log CFU/g in control silages to 4.16 log CFU/g. These results indicate that using LAB as additive can reduce DM losses and improve the microbiological quality of mixed BG and DCP silages.

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Poster 17

Effect of silage from different *Festulolium* cultivars on lamb growth performance

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The efficacy of *Festulolium* in lamb nutrition has not been investigated to date, and the suitability of different hybrids as feed for ruminants has not been evaluated. This study was undertaken to compare the feed value of three *Festulolium* cultivars (Becva – *Festuca arudinacea* x *Lolium multiflorum*, Felopa – *Festuca pratensis* x *Lolium multiflorum*, and Paulita – *Festuca pratensis* x *Lolium multiflorum*), relative to hybrid ryegrass cv. Bakus, in fattening Berrichon du Cher lambs. Experimental silages were made from the first-harvest herbage in their second growing season. Berrichon du Cher rams (24 heads) were divided into four groups, and they were fed an ad libitum experimental diet, for 50 days, composed (DM basis) of: grass silage (60%), ground barley grain (35%), and soybean meal (5%). The lambs were sacrificed at the end of the experimental fattening. The chemical composition of the experimental silages and diets was found. The rams' nutrient intake and growth performance were evaluated. The results were processed statistically by analysis of variance – ANOVA (Statistica 9.0 software, Soft In-corp.). The dry matter content of experimental silages was similar and exceeded 40%. The concentrations of NDF, ADF, and ADL were lowest in silage made from cv. Felopa and highest in silage made from hybrid ryegrass cv. Bakus. Each silage had different WSC content ($P \leq 0.01$). Diets based on silage *Festulolium* cv. Felopa were characterized by the highest intake. Lambs fed this silage had the highest daily and body weight gains. Carcass dressing percentage was comparable in all groups (42.90–43.60%). The feed conversion ratio was lowest in the group fed silage made from *Festulolium* cv. Felopa (3.910 kg DM/kg BW). Funded by the Minister of Science under the „Regional Initiative of Excellence Program”.

Optimizing herbage utilization in dairy sheep with a polyphenol-rich supplement

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One of the problems related to the use of pastures in feeding dairy sheep is the excess of protein or sugar resulting from the intake of grass rich in soluble protein, low in fiber, or rich in water-soluble carbohydrates (WSC). Thus, this work tested a supplement containing a mix of tannins and other polyphenols, formulated by Cargill Inc., to improve feed utilization efficiency in grazing animals. The trial was conducted at a commercial farm in Olmedo (Sardinia, Italy). Forty-eight lactating dairy sheep were selected based on milk production and body weight and fed hay during the night, starchy concentrates at the two daily milkings, and pasture with restricted access time. Ewes were divided into two homogeneous groups and fed, just before grazing, 200 g/day of concentrate without (CNT) or with the additive (ADD). The trial consisted of two sequential periods, the first (11 d) with pasture made by ryegrass (*Lolium multiflorum*), rich in WSC (range WSC: 13.9-36.2% DM; range CP: 9.4-12.7% DM) and then the second (16 d) with pasture made by white clover (*Trifolium repens*), rich in proteins (range WSC: 9.6-12.1% DM; range CP: 17.8-28.2% DM). Milk yield was not affected by the treatment in the whole trial (CNT 1920 g/d, ADD 1923 g/d). However, milk urea was lower in ADD than in CNT (ADD 36.2 mg/dl, CNT 41.4 mg/dl; $P < 0.001$) and milk protein concentration was higher in ADD than in CNT (ADD 5.31%, CNT 5.18%; $P=0.05$) in the first but not in second period. In contrast, milk fat content was higher in the ADD compared to CNT only when clover was grazed (ADD 5.88%, CNT 5.65%; $P=0.004$). When ryegrass was grazed the number of diarrhea cases tended to be higher in the CNT than in ADD ($P < 0.1$). The total count of strongiloides was lower in ADD than in CNT ($P < 0.001$) for the whole experiment. In conclusion, the supplement containing tannins and other polyphenols showed positive effects on the utilization of grass rich in WSC and also when using clover rich in CP, likely due to the stabilization of the ruminal environment, resulting in an improvement in milk composition and health.

Effects of the inclusion of different percentages of red grape pomace on productive factors, milk quality and SCC in dairy goats

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The aim of this experiment was to study the effect of the inclusion of different proportions of the red grape pomace by-product silage in the diet of goats on body weight, individual daily milk production and its macro-composition and SCC. The effect of the inclusion of 0, 6, 12 and 18% of by-product (DM) into isoenergetic and isoproteic diets was studied. 80 goats were selected and distributed into 8 groups of 10 animals each (4 treatments x 2 plots) with homogeneous physiological characteristics. After the pre-experimental sampling, 4 more samplings were done fortnightly: body weight, daily milk production (Lactocorder®, WMB AG, Switzerland), milk composition (fat, crude protein, non-fat dry matter content (NFDm), useful dry matter content (UDM), lactose, urea, MilkoScan FT6000, Foss Denmark) and SCC (Fossomatic 5000, Foss, Denmark) were analysed using a mixed linear model (Proc. Glimmix, SAS v9.4), considering the effect of the covariate in the pre-experimental sampling, the treatment (4 levels), experimental sampling (4 levels) and their interaction (16 levels), and the flock nested to the treatment. The goat was considered the random effect. It was observed that the covariate effect was significant in all variables except for fat and UDM ($P>0.05$). Sampling was significant in all variables except for milk production and lactose ($P>0.05$), while treatment was only significant for urea content ($P<0.05$) where a reduction was observed in the higher inclusion of by-product treatments. An increase in body weight and milk production was observed in the 6% and 12% plots as the experiment progressed, while fat, protein, NFDm, and UDM decreased with the increase in production. SCC did not vary significantly throughout the experiment. It was observed a trend to a production decrease at 12% of inclusion at the end of the experiment, and it was concluded that red wine grape pomace can be included in dairy goat's diet up to 12% DM at short term, and long-term experiments are needed (complete lactation). This study is part of the project PID2021-122962OB-C31, funded by MCIN/AEI/10.13039/501100011033/FEDER, EU.

Baled silage long term conservation: Microbiological and physical-chemical properties

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The use of fermented feeds need to be evaluated due to the risk of Clostridia contamination impairing cheese maturation. This work aimed to evaluate microbiological and physical-chemical parameters of baled silage. A total of 50 samples were collected from 4 farms after 90 days and 180 days from baling and analyzed. Lactic acid bacteria (LAB), mesophilic aerobic bacteria (TMC), yeast, moulds and clostridia in different selective media were counted. Dry matter (DM) content was equal to a mean±SD of 32.1±6.0 and 34.8±6.0% (range 22.4 to 43.4%), while the pH was 3.97±0.16 and 3.98±0.20 (range 3.6 to 4.3), and NDF was 50.9±4.1 and 50.7±3.8% of DM (range 44.2 to 57.5), at 90 and 180 days, respectively. Samples showed sporadically mould (7 over 50 samples, 5 of them at 180 days) never exceeding 10³ CFU/g of silage. The 2.8% of samples was contaminated by clostridia with an average of 330 spores/g of silage (3 samples exceeding 10³ spores/g of silage). LAB, TMC and yeast at 90 days were 6.23, 5.05 and 2.07 log₁₀ CFU/g of silage, respectively. At 180 days of storage, the values of LAB and TMC decreased, whereas yeast slightly increased. Low DM allows higher bale density, better fermentation and lower pH. The low content of ammonia N (4.85±1.9 % Total N) and butyric acid (0.002±0.006 % DM) of baled silage corroborates the recorded low counts of clostridia. A further Principal Component Analysis aggregated the samples per farm, with no differences for sampling time. The first three components explained 65% of the variance. The first component, "Fermentative strength", positively correlated with LAB, lactic acid, ethanol, and 1,2 propanediol. The second component, "nitrogen metabolism", positively correlated with protein and its fractions or N-NH₃, while negatively with sugar content and yeast. The third component "carbohydrate fractions", positively correlated with fiber and acetic acid, while negatively with DM, ADL and starch. Baled silage of high fermentative quality can maintain good quality for dairy sheep feeding until 6 months of storage, with very low Clostridia. Funded by PNRR E.INS ECS 00000038 Spoke 03 APPàre.

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Poster 21

Pasture water-soluble carbohydrates reduced milk fat concentration of lactating ewes by reducing rumen fermentations

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This research investigated the causes of milk fat depression in sheep milk, often observed in late winter-early spring in Sardinian grazing ewes. Five farms of central Sardinia (Italy) were selected. In these farms tank milk and pasture samples were taken weekly for 5 weeks (April 12 to May 7 2021). The supplements used to were also recorded. Milk samples were analyzed for fat, protein, urea and somatic cell count. Milk fatty acids (FA) were analyzed by gaschromatography. Pasture samples were analyzed for CP, ash, crude fat, fiber fractions, and water soluble carbohydrates (WSC). The chemical composition of the pasture (range, DM basis: CP: 6.4- 22.1%; NDF: 36.6-60.4%; WSC: 3.5-26.9%) and of the milk (range: milk fat: 4.40-6.61%; milk protein 5.13-5.81%; fat to protein ratio: 0.80-1.16%) was very variable, based on the farm considered and week of sampling. The data were analyzed to assess possible statistical associations between pasture, milk composition and milk FA. In the first two sampling weeks, milk fat was lower than milk protein (fat to protein ratio: week 1: mean±SD 0.94±0.11%; week 2: 0.96±0.08%), denoting milk fat depression, and then the ratio progressively increased. Milk fat concentration was negatively affected by pasture WSC concentration (milk fat % = 6.536 -0.069 WSC, % DM; r₂=0.47; P<0.01), while it was not associated to other pasture chemical components. Milk fat concentration increased as milk de novo and preformed fatty acid concentration increased, with de novo FA, originated from rumen fermentations, having a much stronger effect than preformed ones (milk fat % = -0.09+3.34 de novo FA % milk, r₂=0.61; milk fat % = 1.80+1.57 preformed FA % milk, r₂=0.87). Milk fat concentration was also inversely associated to the de novo to preformed FA ratio (r=0.69) and to the branched chain milk FA (r =0.73), which are indicatory of rumen microbial activity. Milk vaccenic (C18:1t11) and rumenic (CLAc9,t11) acids were highly associated (r =0.96) between them, but not to milk fat concentration. In conclusion, it appears that the milk fat depression associated to high pasture WSC is due to a marked reduction of microbial fermentations in the rumen.

Seasonal nutritional values of *Rubus fruticosus* leaves as a sustainable alternative feed for goat in mountain regions

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Rubus fruticosus leaves, renowned for their medicinal and veterinary uses, have been insufficiently studied for their nutritional value. This study investigates the chemical composition and ruminal degradability of *Rubus fruticosus* leaves in goats and across seasons. *Rubus fruticosus* leaves were collected from two valleys in the NW Italian Alps at an altitude of 700m, throughout the year (3 samplings/valley/season). Chemical analysis and in vitro tests to measure true and apparent dry matter degradability (TDMD, ADMD), and fiber degradability (NDFD) at 48h were conducted using rumen fluid collected from slaughtered goats. The results revealed the potential of *Rubus fruticosus* leaves as a promising alternative protein feed with high-fiber compound, but limited NDFD. Seasonal variability significantly affects moisture, crude protein content, and degradability. Spring emerged as the optimal season with the highest nutritional value, including the high crude protein content (21.6% dry matter, DM), moisture (76.2% of fresh matter), ADMD (50.3% DM), TDMD (73.2% DM) and NDFD (25.0% neutral detergent fiber, NDF). Conversely, mineral and fiber compounds, such as NDF, acid detergent fiber, and lignin, remained relatively stable across seasons, averaging 6.3, 37.5, 22.4 and 8.4%, respectively. In conclusion, *Rubus fruticosus* leaves offer a valuable and sustainable alternative feed for goats, particularly in mountain regions, especially during spring. This presents an opportunity to diversify feed resources, reduce reliance on conventional feeds, mitigate the impact of feed shortages in mountain areas and improve the sustainability of the mountain goat production systems.

Session 82

Poster 23

In vitro ruminal fermentation of dairy diets including dried brewers' grains

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The aim of this study was to assess the in vitro ruminal fermentation of dairy ruminant diets including increasing amounts of dried brewers' grains (DBG). The DBG sample contained (dry matter (DM) basis): 3.61% ash, 26.6% crude protein (CP), and 65.8% neutral detergent fiber (NDF). Four isonitrogenous and isoenergetic diets were formulated: a control diet without DBG, and 3 diets including 6, 12 and 18% of DBG (fresh matter basis). The control diet contained 45% of alfalfa hay, and in the other diets the DBG partially substituted corn DDGS (dried distillers grains with solubles), soybean meal and alfalfa hay. Diets were fermented in vitro with buffered sheep ruminal fluid (4 replicates) for 24 h to determine rumen fermentation parameters (pH, volatile fatty acids (VFA) and NH₃-N) and gas production was measured at 4, 8 and 24 h. Gas production at 4, 8 and 24 h decreased linearly ($P \leq 0.003$) with increasing DBG inclusion, which is consistent with the greater gas production observed for alfalfa hay, corn DDGS and soybean meal compared with DBG when each feed was incubated individually. However, there were no differences ($P > 0.05$) between diets in final pH and total VFA production, although propionate proportion decreased ($P = 0.045$) and acetate/propionate ratio linearly increased ($P = 0.044$) by increasing DBG inclusion. Concentration of NH₃-N at 8 h of incubation decreased linearly ($P = 0.011$) as the amount of DBG in the diet augmented, but no effects were observed after 24 h of fermentation. This could be due to a slower CP degradability of DBG compared with the replaced feeds. In conclusion, DBG can be included up to 12% in diets for dairy ruminants without negative effects on ruminal fermentation.

Variability in chemical composition of barley rootlets from different malting plants

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Barley rootlets (BR) are the dried shoots and rootlets of sprouted grain generated in the malting process previous to beer brewing process. Although BR are used in ruminant feeding, information about their chemical composition is limited. This work aimed to analyze the variability in chemical composition of 10 samples of BR obtained from 3 Spanish malting plants in different periods. Samples were ground to pass a 1 mm screen and analyzed for dry matter (DM), ash, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ether extract (EE). The DM content ranged from 94.7 to 96.7% (average 95.6%) and the ash content ranged from 5.43 to 7.10% (average 6.14%), whereas CP ranged from 25.1 to 39.3%, with average value of 31.4% (all values on DM basis). The average contents of NDF, ADF and EE were 48.6 (41.7 – 56.0%), 15.7 (11.4 – 20.5%) and 2.27 (1.65 – 2.75%), respectively. Coefficient of variation was high for CP (12.1%), ADF (15.2%) and EE (14.1%), but low for DM (0.62%). Both the DM and CP values of our samples were higher than those reported by Feedipedia ranging from 86.0 to 94.9% for DM (average 89.9%) and from 17.3 to 30.7 for CP (average 23.5% of DM). In contrast, ash, NDF, ADF and EE contents were similar to those in INRA-CIRAD-AFZ Tables (average 5.9, 44.6, 16.5 and 1.8% of DM, respectively). A negative linear correlation was observed between NDF and CP content ($r = -0.633$; $P = 0.049$). The results indicate high variability in chemical composition of BR showing differences in CP, ADF and EE content compared with available data. These variations may be due to differences in the composition of the barley grains used in different malting batches, as well as in the malting process performed at each malting plant.

Session 82

Poster 25

Ruminal fermentation and biohydrogenation in ewes fed with sainfoin pellets under water restriction

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Sainfoin (*Onobrychis viciifolia*), a Mediterranean legume forage, has a great interest as feed due to its content of condensed tannins (CT), which are known to be able to interfere in ruminal processes, affecting fermentation and lipid metabolism. The aim of this study was to characterize the rumen fermentation and biohydrogenation in two groups of adult Rasa Aragonesa ewes (60 ± 2.7 kg BW), under water restriction and parasitized, fed with straw plus 300 g of cereal-concentrate ($n=20$; CONTROL) or 300 gr of sainfoin pellets ($n=20$; SAINFOIN) with 16.5 g CT/kg DM. The trial lasted 8 weeks, 4 of which under 40% water restriction. After that, the ewes were slaughtered and their ruminal content was extracted to analyse pH, volatile fatty acids (VFA), and fatty acid (FA) profile in the rumen. The SAINFOIN diet reduced the total VFA and the proportions butyric, isobutyric, valeric, and isovaleric acid ($P<0.05$), increased the proportion of propionic acid ($P<0.01$), and tended to increase the production of acetic acid ($P<0.10$). Regarding the fatty acid profile, SAINFOIN ewes presented lower proportions of C18:0 and trans-monounsaturated FA ($P<0.05$), suggesting a reduction of the ruminal biohydrogenation caused by the presence of sainfoin CT. Besides, SAINFOIN diet increased the proportion of polyunsaturated fatty acids (PUFA) n3 ($P<0.001$), that could enhance the immune function, greatly reducing the PUFA n6/n3, and also led to a higher total conjugated linoleic acid percentage (CLA; $P<0.001$) and its main FA, CLA c9t11. Although sainfoin supplementation reduced total VFA, the many findings in the ruminal FAs suggest promising improvements concerning the ruminal FA metabolism that could help these ewes to cope with different stresses.

Effects of protein sources and microbial addition to potato waste on silage fermentation and in vitro digestibility

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This study evaluated effects of protein sources (soybean meal and amarula nut oil cake) and microbial addition on the nutritive value, fermentation characteristics, aerobic stability and in vitro digestibility of ensiled potato wastes (PW). A quantity of 210 g PW/kg fresh matter (FM) was mixed with 450 g grape pomace (GP)/ kg FM plus 150 g sugarcane molasses/kg FM and 300 g amarula nut cake (ANC). This mixture was treated either without (T1) or with *Lactobacillus planturum*, (LAB, T2). The other batch replaced ANC with 300 g soybean meal (SBM), and was treated either without (T3) or with LAB (T4). The last batch contained 150 g ANC mixed with 150 g SBM, treated either without (T5) or LAB (T6), making 6 silages in total. The mixtures were ensiled in 1.5 L jars and were kept for 60 days in a room of 27°C. Samples were collected on days 0 and 60 of ensiling for the determination of fermentation indices and in vitro nutrient digestibility. The addition of LAB improved ($P<0.05$) silage fermentation but did not affect the aerobic stability of the silage. Addition of SBM improved ($P<0.05$) the protein (CP) and dry matter (DM) of the silage while the addition of ANC increased ($P<0.05$) both the fat and fibre of the silage. However, the fibre (NDF and hemicellulose) was reduced ($P<0.05$) in silage that contains the mixture of SBM and ANC. These combinations also improved ($P<0.05$) the digestion of the hemicellulose and microbial yield of the silage. Potato waste silage produced with the combination of ANC and SBM yielded feed that is rich in nutrients, and should be fed to ruminants to measure animal growth performance.

Session 82

Poster 27

Performance from birth to slaughter of single and twins Merilin Plus® lambs under native grassland conditions

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Merilin Plus (MPlus) is a maternal breed born in Uruguay ten years ago from crossing of Merino x Finnish rams and Merilin ewes. Little information in reproductive and productive performance of this breed is available. The aim was to report comparative performance of simple and twin lambs from birth to weaning/slaughter (80 days old). During breeding season (32° S 58° W; Uruguay) 44 multiparous Mplus ewes (53.7±4.9 kg liveweight -LW-, and 3.5 years old) were mating with one Mplus ram. Under native grassland pasture conditions ewes were supplemented at peri-partum with whole barley (0.5% of LW/day). Sixty six percent of ewes lambing single lambs and 32% of them twins. From 50 day-age, all lambs received during 30 d (creep feeding system) a barley grain and soybean meal mixture (70/30%; 1% of LW/day; 2.9 Mcal EM and 22% CP). At birth, twin lambs (n=28) were lighter than single lambs (n=29) (4.4±0.7 kg vs. 5.2±0.7 kg; $P<0.05$), without differences in lamb survival (100 vs. 100%). At weaning, twin lambs were lighter (21.5 vs. 26.8 kg), gain less weight (0.220 vs. 0.285 kg/day; $P<0.05$), and had less body condition (3.3 vs. 3.6) than single lambs. However, twin ewes produced more kg of LW/kg ewe mating than single ewes (0.80 vs. 0.52 kg; $P<0.05$). In the slaughterhouse, twin lambs (n=17) had lower carcass weight (9.6 vs. 12.6 kg) and carcass dressing (48.2 vs. 50.7%), worse carcass conformation (6 vs. 0% Inferior; 76 vs. 22% Medium; 18 vs. 78% Prime), and lower GR point (3.9 vs. 6.3 mm) than single lambs (n=18). Nevertheless, twin ewes produced more kg of carcass weight/kg ewe mating than single ewes (0.37 vs. 0.24 kg; $P<0.05$). We concluded that twins had lower performance at 80 day-age than simple Mplus lambs, but twin Mplus ewes were more efficient.

Effect of early life nutrition on the growth, metabolic and carcass characteristics of beef cattle

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Early adaptation to a perinatal nutritional stimulus permanently alters cattle physiology and metabolism, significantly impacting key productivity and longevity traits such as reproductive development, pubertal advancement, and lifetime milk production. In beef production systems, maximizing carcass gain, achieving high meat yield, and meeting industry specifications for desirable quality attributes are crucial economic traits. Our research shows that up to 40% of the variation in final carcass weight is attributable to calf performance during the first three months of life. Failure to meet early growth targets by weaning and housing (under 7-8 months of age) often results in lighter carcasses and/or delayed age at slaughter, adversely affecting production efficiency, farm economics, and environmental impact. Enhanced early-life nutrition in beef calves has been shown to positively influence lifetime growth potential and carcass composition, but data on this are limited. Key developmental programming windows in early calf life are proposed to influence carcass composition and development. Strategic nutritional manipulation during these windows can enhance tissue hyperplasia and hypertrophy, particularly through targeting myogenesis—via the post-natal division of skeletal muscle satellite cells—and adipocyte development, characterized by hyperplastic growth. This approach may alter carcass gain, adiposity, composition, and meat quality later in life. This presentation will explore the effects of early-life growth and targeted nutritional strategies on tissue development at physiological and molecular levels, and will address the immediate and latent impacts on animal performance, carcass gain, composition, and production and environmental efficiency within sustainable pasture-based dairy beef systems.

Session 83

Theatre 2

Effects of undernutrition and hydroxytyrosol supplementation during the last third of gestation on growth and physiological profiles of male beef offspring

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Effects of maternal subnutrition and hydroxytyrosol (HT, antioxidant from olive leaves) inclusion during late pregnancy on live weight (LW), average daily gain (ADG) and physiological profiles were studied in male beef offspring during the fattening period (4 to 12 months of age). From gestation week 28 to calving (w40), 109 cows were divided into four groups: feeding level (100 vs 60% requirement) x HT (Control vs HT, for 0 and 178 mg HT/kg unifeed). Parda de Montaña (n=25) and Pirenaica (n=21) concentrate-fed bulls were weighed monthly, and blood samples were collected bi-monthly to determine plasma levels of glucose, fructosamine, urea, creatinine and IGF-1. Data were analysed with a mixed linear model with feeding, HT, time and breed as fixed effects, and bull as random effect. Live weight at the end of the fattening and ADG during the fattening period were not affected by maternal feeding level (496 vs 498 kg LW; 1.18 vs 1.40 kg ADG; for 100 and 60%), HT inclusion (493 vs 501 kg LW; 1.22 vs 1.36 kg ADG; for Control and HT) or breed (501 vs 493 kg LW; 1.23 vs 1.35 kg ADG; for Parda and Pirenaica). Concerning physiological profiles, urea was the only one affected by the interaction feeding x HT (at 10 and 12 months of age). Interestingly, fructosamine and glucose levels did not follow a similar evolution over time. To sum up, maternal subnutrition and HT during the last third of gestation had no major effect on male offspring growth and physiology during the fattening phase. More studies are needed to assess their potential effects on male meat quality. Funded by PID2020-113617RR-C21 FETALNUT. Research group A25-23R.

Changes in the plasma metabolome of Holstein heifers from birth to first insemination

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The objective of this study was to characterise the plasma metabolome in Holstein dairy heifers from birth throughout the rearing period until the first insemination, including the effects of the dietary changes related to weaning. Blood samples were collected from 30 Holstein-Friesian calves at eight key time points during the rearing period, i.e., six times during calthood (birth until the end of weaning at 98 d of life) and then at month 8 and 13. All calves received 4 L of colostrum after birth and were fed 12 L of milk replacer per day (140 g/L) until the start of gradual weaning (d 57 to 98 of life). All plasma samples were subjected to a targeted metabolomic analysis (MxP® Quant 500 kit of Biocrates Life Science AG) with liquid chromatography-tandem mass spectrometry. In total, 243 plasma metabolites were identified and were categorised according to compound class. Across all time points, glucose, amino acids (n=20), carboxylic acids (n=2), cholesterol esters (n=15) and glycerophospholipids (n=73) were the most prevalent metabolite classes. Principal component analysis (PCA) showed differences in metabolic profiles between the sampling time points. As the calves matured, the concentrations of lipids (glycerophospholipids, sphingolipids and triacylglycerols) increased significantly indicating an increase in the rate of fatty acid metabolism to promote the synthesis of cell membranes, hormones and other molecules required for development. Overall, our results provide detailed insights into the metabolic changes during the first year of life in young heifers.

Session 83

Theatre 4

Serum lipidome profiling of dairy calves fed high-fat milk replacers containing animal or vegetable fats

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This study aimed to investigate the serum lipid profile of individually housed calves. Thirty male Holstein calves (mean weight 45.7 kg; age 2.4 d) were randomly assigned to 2 diets (n = 15/group) consisting of 1) a MR with only vegetable fats containing 40% Racomelt and 60% rapeseed oil (VG); 2) a MR with animal fats containing 65% packer lard and 35% dairy cream (AN). Both MR contained 30% fat, 24% crude protein and 36% lactose, fed to calves in two equal meals via teat buckets at 135 g/L. At 35 d after arrival, blood samples were collected and analysed by untargeted lipidomic LC-QTOF-MS to determine lipid profiles. PCA and OPLS-DA revealed distinct lipid profile (594 species, 25 classes) between VG and AN group, reflecting varied metabolic responses to MR fat composition. Calves fed AN had 39 higher and 171 lower lipid species than VG-fed calves (Volcano plot, FC ≥ 1.5, FDR ≤ 0.05). Lipid classes including phosphatidylcholine (PC), phosphatidylethanolamine (PE), sphingomyelin (SM), triglycerides (TG), lysophosphatidylcholine (LPC) and lyso-PE (LPE) differed significantly between treatments. Lower PC and PE concentrations in AN-fed calves suggested potential alterations in cell membrane integrity, intracellular signalling pathways, and hepatic function. The lower levels of certain TG types in AN-fed calves could be due to differences in the fatty acid profiles between VG and AN. Lower levels of phosphatidylinositol (PI), LPC, LPE, lyso-PI and SM species in AN-fed calves also indicate changes in lipoprotein and lipid metabolic pathways. These results deepen our understanding of how MR fat composition affects calf serum lipidome profiles.

Phenotyping the trans-10 shift in the rumen of finishing lambs

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The microbial community in the rumen is responsible for metabolizing unsaturated dietary lipids through biochemical pathways known as biohydrogenation. Among these pathways, the normal pathway produces t11-18:1 as the principal product, while the altered pathway (t10-shift) generates t10-18:1. There is still little knowledge about the factors that drive the t10-shift and the variability in susceptibility among animals. This study aimed to phenotyping the variability of t10-shift expression in finishing lambs. For that, over 1.5 years, 383 lambs from 7 genotypes and different regions were finished in the same farm. At slaughterhouse rumen contents were collected and later fatty acids were analyzed from the freeze-dried sample. The results showed that the proportions of t10-18:1 ranged from 0.36-27.15%, and t11-18:1 ranged from 0.22-17.01%. A large number of animals were in shift (t10/t11>1), with only 13% of animals presenting a shift <1, and an impressive 62% of animals showed a shift >5. There were noticeable differences in t10/t11 among genotypes, with Romane showing the lowest (3.4) and crossed-Merino the highest average (10.8). Further studies are needed to evaluate whether genotype is critical for t10-shift resistance. Financial support was provided by PTDC/CAL-ZOO/4515/2021, UIDB/152816/2022, UIDB/00276/2020 and LA/P/0059/2020 projects.

Session 83

Theatre 6

A common database to study the efficiency of the mother-young pair in suckling animals

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The efficiency of the mother-young pair corresponds to the mother's capacity to produce a healthy and well-growing young and to maintain its production. The aim of this work was to create a common database to explore the efficiency of the mother-young pair through generic indicators in three suckling species: cattle, ewes and horses. The database regroups 604 births for beef (from 2000 to 2022), 6345 births for ewe (from 1999 to 2014) and 462 births for horse (from 2010 to 2022). Within each species the mother young pairs were identified per year as mothers can be studied over several years as they have more than one young during their productive life. The following performances for at least one specie were collected: milk production, weight of the mother (before and after parturition), weight of the young (from birth to weaning), average daily gain of the young (from birth to weaning), body condition score of the mother (at the beginning of the reproductive season, after weaning) and body measurements of the young (chest girth, withers height and canons width from birth to weaning). Other information such as the breed; parity, number of lactations; age of the mother; birth and weaning dates of the young were recorded. A large number of these variables can be influenced by animal (e.g. genetics), nutritional factors (e.g. feed provided to both units of the pair) and management system (e.g. age at parturition, parturition's period). Initial relationships between variables were established to identify generic proxies for mother-young pair efficiency between the three suckling species. One of the studied proxy is the live weight of the young/litter (at birth and/or at weaning) divided by the live weight of the mother at parturition helping us to determine how the different performances are correlating with this indicator.

Incorporation of butyric and caproic acids into milk replacers for calves fed twice daily

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Milk replacers (MR) contain low levels of butyric (C4:0) and caproic (C6:0) acids because of the use of alternative fat sources to milk fat. This study examined how the incorporation of tributyrin (TB) and tricaproin (TC) into MR affects the intake, growth, and health of calves. Sixty Holstein male calves (18.4 ± 2.4 d) were blocked by BW and within each block, calves were randomly assigned to 1 of 4 treatments (15 calves each): 1) a control MR with vegetable fats (CON), 2) CON with TB, 3) CON with TC, and 4) CON with both TB and TC (TBTC). All MR were isoenergetic with 27% fat, 24% CP, and 36% lactose (DM basis). The MR allowance was 7L/d (13.5% solids) fed in 2 meals. Starter feed, straw, and water were provided ad libitum until 63-d old. Calves were individually housed. Body measurements and BW were measured weekly. Feed intakes and fecal consistency were recorded daily. Continuous variables were evaluated as repeated measures and discrete variables were analyzed using mixed effect logistic regression. A time and treatment interaction (T×P) showed lower wither height at 7 wk ($P=0.01$) and lower body barrel at 7 and 8 wk after arrival ($P=0.02$), in calves fed TBTC and TB than other treatments, respectively. A T×P interaction and a trend indicated that MR intake of TBTC ($P=0.01$) and TC ($P=0.05$), respectively, was lower compared to other treatments, with increased MR refusal rates up to 5 wk after arrival. During the entire period, CON group had a higher percentage of days with loose feces ($P<0.05$) and more therapeutic interventions for fever and respiratory disease than other treatments ($P<0.05$). Incorporating both TB and TC in MR fed to 2-wk old calves increased MR refusals but reduced both therapeutic interventions and diarrhea incidence.

Session 83

Theatre 8

Incorporation of butyric and caproic acids into milk replacer fed to newborn calves

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Milk replacers (MR) contain low levels of butyric (C4:0) and caproic (C6:0) acids because of the use of alternative fat sources to milk fat. This study evaluated how the incorporation of TB and TC in MR affects the intake, growth, and digestive health of newborn Holstein male calves. Twenty-four calves (0.90 ± 0.35 d old) were blocked by arrival day and age. Within each block, calves were randomly assigned to 1 of 4 treatments (6 calves each) of different ratios of a control MR mixed with a MR containing TB and TC at the inclusion levels of 0, 33, 67, and 100%. All MR were isoenergetic with 27% fat, 24% CP, and 36% lactose (DM basis). The MR allowance was 7 L/d (15% solids) fed in 2 meals. Daily MR intake and drinking assistance were measured by weighing refusals 10 and 30 min after feeding. Starter feed was unavailable; however, water and straw were provided ad libitum. Calves were housed individually and weighed weekly. Fecal consistency was scored daily for 21 days. Continuous variables were evaluated as repeated measures and discrete variables were analyzed using mixed effect logistic regression. Growth and MR intake were not affected by the treatments ($P>0.05$). However, calves fed level 0 required more drinking assistance ($P=0.01$); conversely, higher TB and TC levels in the MR led to a reduced need for drinking assistance. In the first wk of life, fecal scores did not differ across treatments. However, calves fed levels 0 and 33% in wk 2 ($P=0.05$) and 3 ($P=0.09$), tended to have a higher proportion of abnormal fecal scores than the other treatments. Incorporating TB and TC into MR did not affect intake and even reduced the need for drinking assistance in newborn calves. Additionally, it reduced the incidence of diarrhea.

Dietary lysine and protein for organic grower-finishing pigs

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The objective of the study was to determine the physiologically optimal supply of standardized ileal digestibility (SID) of lysine for organic growing-finishing pigs. In total, 150 pigs (LY-TN70 x Danbred-Duroc) were included in the experiment from 31 kg (\pm 4.3 kg) until slaughter at 112 kg (\pm 11.0 kg). Initially, pigs were distributed according to BW and sex in 15 pens with 10 pigs in each. Pigs were allotted to five different treatments with increasing SID lysine content of 6.1, 6.5, 6.9, 7.3, and 7.7, g/kg, SID CP increased accordingly. The AA ratio relative to lysine and physiological energy in diets remained constant across dietary treatments. Pigs were fed ad libitum with grass clover silage (GCS). Feed and GCS intake for each pen was recorded daily. Pigs were weighed and blood samples were taken initially and at slaughter. At slaughter, hot carcass weight and meat percent were measured. Data were analyzed using the MIXED procedure of SAS. Linear and quadratic effects of dietary treatments were tested. P-values were adjusted using a Tukey test, to account for multiple comparisons. Increased dietary SID lysine did not affect pen average daily feed intake (ADFI; $P = 0.90$), pen GCS intake ($P = 0.94$), pig average daily gain (ADG; $P = 0.76$), pen feed conversion ratio (FCR; $P = 0.12$), days to slaughter ($P = 0.75$), nor hot carcass weight ($P = 0.61$). Pen FCR tended to decrease linearly ($P = 0.05$), with increasing dietary SID lysine (2.76 to 2.64 kg feed/kg gain), whereas meat percent increased linearly ($P = 0.01$) with increasing dietary SID lysine (58.6 to 59.8%). In conclusion, diets for organic grower-finisher pigs could be reduced to 6.1 g SID lysine/kg feed without affecting ADG and ADFI, however, FCR and meat percent may be compromised.

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Theatre 1

Precision Livestock Farming technologies could reduce greenhouse gas emissions on beef farms

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To achieve their net zero by 2045 target, the Scottish Government have called for a 31% reduction in agricultural emissions by 2032 from 2018 levels. Recently, precision livestock farming (PLF) technologies have been highlighted as a promising strategy to indirectly reduce greenhouse gas (GHG) emissions through increasing production efficiencies. Utilising data from the Scottish Cattle Tracing System, we created two baseline scenarios (one grazing and one housed system) and modelled the effects of various PLF technologies on whole farm and product emissions. Baseline emissions were calculated using the Agrecalc carbon footprinting tool. Scenarios modelled included adoption of an automatic weigh platform, accelerometer-based collars for oestrus detection and accelerometer-based collars for early disease detection. All three PLF technologies reduced total emissions and product emissions in both the grazing and housed systems. The largest reduction in total emissions were seen following the adoption of an automatic weight platform which reduced the age of slaughter by three months in the grazing system (6.8%) and following the adoption of health collars in the housed system (6.1%). Health collars also resulted in the largest reduction in product emissions for both the housed (12.0%) and grazing systems (10.5%). Our modelling suggests that PLF technologies could be an effective GHG mitigation strategy for both grazing and housed beef systems. Although this study was based on data from beef farms in Scotland, comparable emission reductions are likely attainable in other European countries with similar farming systems.

PLF-assisted quantification of carbon footprint; Preliminary analysis of Agroforestry system

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The majority of farms in the EU (>80%) are small-scale operations spanning 1 to 20 hectares, characterized by limited productivity and facing economic challenges (a 30% decrease since 2010). For these farms, agroforestry practices, alongside the emerging market of Carbon Credits, present new opportunities for value creation. This study aims to evaluate the feasibility of transitioning from costly manual carbon certification protocols to a digital model based on Precision Livestock Farming (PLF) and Precision Agriculture (PA) systems. The experimental site, spanning 3 hectares in Abruzzo, Italy, implemented a Silvopastoral production system that combined rotational grazing of broiler chickens (200 animals) with hazelnut cultivation (650 trees). A unified Life Cycle Assessment (LCA) model was developed to establish a comprehensive carbon balance. Poultry weight was monitored using a Walk-Over-Weight (WOW) system, while vegetation growth dynamics were assessed using satellite spectrum analysis (AGB and LAI, Sentinel-2). The production setup incurred a carbon cost of 2.73 tons of CO₂ equivalent, with emissions from poultry production amounting to 5.52 tons of CO₂ equivalent per year. The sequestration potential was estimated at 1.8 tons of CO₂ equivalent per hectare of pasture and 5 kg of CO₂ equivalent per year per tree, resulting in a total sequestration of 8.65 tons of CO₂ equivalent per year. The WOW system attributed an average of 45 kg of CO₂ equivalent per animal, while satellite imaging detected vegetation dynamics, indicating a dry matter increase from 5 tons/ha in low seasons to 13 tons/ha in high seasons. This data was consistent with manual “Gold standard” measurements conducted four times during the animal production cycle and twice a year for plant dynamics. In conclusion, Silvopastoralism, combined with on-site LCA modeling, can provide farmers with access to the emerging Carbon Market, with the integration of PLF and PA being able to significantly reduce costs. However, some on-site measurements are still required, alongside a more widespread integration of technology and digital literacy among farmers.

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Theatre 3

Projection of Thermal Humidity Index (THI) and Frequency of Severe Heat Events for Broilers and Layers in the Context of Climate Change in Brittany, France

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The intensification of climate change poses significant challenges to the poultry sector, particularly in regions like Brittany, which produces one out of every three broilers and 36% of the consumption eggs in France. Rising temperatures can detrimentally affect the thermal comfort and physiological well-being of broilers and layers as well as decrease the overall productivity. To anticipate the impacts of climate change on poultry production, a simulation-based study to project the Thermal Humidity Index (THI) and the number of days exceeding a severe threshold outside and within animal houses has been conducted. Utilizing comprehensive French climate simulations and a computational model based on thermodynamic equilibrium, factors such as animal density, airflow rates, and misting levels were incorporated to simulate outdoor and indoor weather conditions representative of broiler and layer housing systems. The results revealed an increase in THI values across broiler and layer farms in Brittany, with a corresponding escalation in the frequency of days exceeding the severe heat stress threshold. These projections underscore the urgent need for adaptation measures within the poultry sector to mitigate the adverse impacts of climate change. The developed model can be used as a tool to investigate such measures by optimizing the combination of ventilation systems, stocking densities, and misting strategies in the case of heat stress. Through the integration of these adaptation strategies, the poultry sector can investigate the challenges presented by climate change and enhance the resilience of poultry production systems in Brittany and beyond.

Coupling precision livestock farming technologies with artificial intelligence for mitigating heat stress in Mediterranean dairy cattle systems

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Rising temperatures in Mediterranean regions represent not only the risk of heat stress and deterioration of animal welfare but also cause a negative impact on the productivity of dairy production systems (DPS). In this context, facilitating the adoption and evaluation of context-specific strategies to alleviate heat stress, particularly in areas such as the Mediterranean that suffer the most from rising temperatures, is paramount. To this end, this study proposes two integrative strategies combining precision livestock farming (PLF) technologies and artificial intelligence (AI) to mitigate heat stress. The first approach focuses on shading in outdoor feedlots as a strategy to mitigate heat stress. The temperature-humidity threshold (THI) at which animals seek shaded areas was identified using PLF tools, such as cameras and machine learning algorithms. Preliminary results allow us to indicate which is the combination of day/night THI values above which animals come to the shade, thus allowing optimisation of shade management to improve animal welfare. The second approach identified the optimal showering regimes in DPS by combining AI and collars for behavioural monitoring. The results show that increased daily showers can significantly mitigate heat stress in dry and lactating cows, reducing panting by up to 36%. In all, these approaches offer knowledge-based solutions for farmers, allowing the optimisation of their management practices and ensuring animal welfare and the long-term sustainability of the DPS across Mediterranean environments.

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Theatre 5

Hot topic: novel weather features to quantify heat stress in dairy cows in moderate climatic conditions

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Heat stress has been a 'hot topic' in the past few years, as the weather has become more volatile and extreme in the wake of climate change. Previous research on heat stress in dairy cows mainly focused on describing changes in milk production or behavior as a function of fixed day-average temperature-humidity index (THI) classes. However, in the moderate climatic conditions of Western Europe, severe heat stress with THI above 75 is rare, yet heat stress consolidates in longer moderate-high THI periods without an opportunity to cool down. Therefore, with the previous approach, the quantification of the heat stress sensitivity of animals is often estimated based on a limited amount of data. Our study aims to quantify the effect of outdoor climatic conditions on cows held in intensive production systems. More specifically, the level and impact of heat stress depend on the duration and severity of the insults and the capacity of the animals to recover e.g., during the night. This explorative study presents a several weather features designed to capture the outdoor climatic conditions relevant to dairy cows heat stress. They include the severity and duration of the temperature or THI insults and the time-lagged and continued effects of the hot weather insults. We quantified these features as calculated from hourly weather data of Belgium, the North of France, East Germany and the Netherlands for a period of 19 years (2005-2023) and link them to responses in milk production and step count of dairy cows.

Variation among individual beef cattle in methane measurements with laser methane detector

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Laser methane detector (LMD) for classification of high and low emitters is becoming widespread as an expeditious method at sustainable costs. However, to date, there is still little evidence on beef cattle. The aim of this trial was to investigate whether specific problems exist and evaluate the main factors influencing phenotype variability. CH₄ emissions of 10 beef calves, housed in five-place pens, were recorded for two consecutive days using LMD. Two operators (emitter distance between 1 and 4 m) recorded the calves from 1' to 5' based on the position and animal activity. In total, 247 individual measurements (13-40 per calf) for a total of 67434 data points were produced. Three types of CH₄ phenotype were considered (ppm*m, log-ppm*m and sum-ppm*m). The CH₄ variables were adjusted for the fixed effect of age groups, behavior (standing, laying down-ruminating, feeding) and operators. The association among standardized residuals of statistical model were assessed by spearman rank correlation (r). The emissions were significantly and constantly different among behaviors: laying down-ruminating>feeding>standing (p<0.001). However, emitters were classified with different rankings according to variable used (r=0.80 ppm*m vs log-ppm*m, r=0.68 log-ppm*m vs sum-ppm*m and r=0.83 ppm*m vs sum-ppm*m). This preliminary analysis showed the LMD were useful for detecting individual methane emissions also in beef cattle, but different emission rankings were observed according to measurements. Similarly to what observed in dairy cattle methane monitoring by LMD, for animals housed in pens it seems necessary to validate a specific measurement routine.

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Theatre 7

Towards automated monitoring of ammonia emissions from naturally ventilated pig houses

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The developments of gas sensors and communication technologies in the recent years is opening an opportunity to conduct real-time measurements on naturally ventilated commercial farms. A commercial fattening pig farm located in Aragon (Spain) was monitored. The farm had two identical buildings (Building 1 and Building 2) with a capacity of 980 animals each. In each building, 6 measurement locations were selected where both ammonia sensors (DPF-Q500D, DPF Sensors) and carbon dioxide sensors (CO₂-42-DIS, DPF Sensors) were placed. Temperature and relative humidity were also measured. Additionally, two external gas sensors were installed. All information was registered every minute and real-time stored in a cloud system that could be accessed remotely. The measurements were conducted during one fattening cycle starting in Summer 2023 and a second cycle has started in Winter 2024 (results of the second cycle are not yet available). Ammonia emissions were calculated on an hourly basis following a mass balance, where ventilation rate was estimated using a CO₂ balance. The CO₂ balance used the CIGR recommendations to calculate energy and carbon dioxide production from the animals, as well as for the temperature and animal activity corrections, while the emission from the slurry was considered negligible. Emissions could be calculated on 94% of measured hours. The remaining 6% hours were neglected due to insufficient difference between indoor and outdoor CO₂ concentrations. Emissions followed a curve that responded to animal growth and slurry removal events. On average, emissions were 0.73 and 1.23 kg/place/year. Results show a potential to calculate emissions automatically, in a process that could be improved using precision livestock farming techniques to improve the animal weight estimation and daily activity patterns required to operate the CO₂ balance. These results are part of the R+D+i project CPP2021-008476, funded by MCIN/AEI/10.13039/501100011033/ and by the "European Union NextGenerationEU/PRTR"

Estimation of NH₃ emission factors for pig fattening buildings thanks to Machine Learning methods and ELFE database

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In a project called PigAmmo, the aim of this study was to estimate NH₃ emission factors in pig production by using Machine Learning methods based on the use of the database ELFE. ELFE is a database gathering emission factors available in the literature and its metadata concerning housing and storage conditions, and measurement methodologies (Vigan et al. 2019). After an initial exploration of data, the work focused on ammonia emissions of conventional fattening units with forced ventilation. The Machine Learning models were created in Python thanks to Scikit-learn package (Pedregosa et al. 2011). The first step was to clean up the database so that only emission factors almost completely filled were retained. We also deleted observations where specific protocol can modify ammonia emission like feed additives. After this, from 590 observations describe by 32 features, only 20 features about 472 observations were kept. When data of interest still missing, they were filled in by expert opinion and/or automatic calculation: median value for numerical feature and the most frequent modality for categorical feature. Several algorithms were tested to compare and choose t, based on coefficient of determination (r^2) between real and predicted values. Although the best training performance reached a high value ($r^2=0.92$), the test database score decreased to an unacceptable value ($r^2=0.60$) for predicting reliable NH₃ emission. Heterogeneity and lack of fully completed data in the ELFE database explained these disappointing results and illustrate the importance of metadata into scientific publications. Vigan et al. 2019, doi: 10.2134/jeq2019.01.0007 ; Pedregosa et al. 2011, doi: 10.5555/1953048.2078195

Designing a thermoregulated unit for studying the impact of temperature on zootechnical and environmental performance of growing pigs

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As part of an ADEME-funded project to study the environmental impact of temperatures on gaseous emissions during pig fattening, a thermoregulated unit called ClimatoTec has been built at the Ifip experimental station in Romillé (France). This unit is composed of two rooms, each with a capacity of 20 pigs between 25 and 118 kg. Each room is divided into two pens housing 10 pigs each, raised on fully concrete slatted floors. The slurry is stored in a pre-pit throughout the animals' stay. The air supply in each room is provided by a Flud'R with 0.15m², while the extraction uses a 350mm fan located underneath the slatted floor. These facilities are each equipped with a reversible heat pump, a cooling or heating device depending on the type of test carried out (hot or cold conditions), a water tank and an air heater. The air heater is responsible for drawing outside air through a water battery before entering the attic of the room. The battery is fed by the water from the storage tank so that the air in contact with it can cool or warm up without altering its absolute humidity. The air heater features a fan with a progressive operating mode controlled by the room's regulating unit. The heat pump enables the circulation of the refrigerant fluid in a double circuit, one directed towards the tank and the other towards the device for dissipation or heat recovery (depending on the type of test). This new installation provides a valuable resource for studying the effects of climate change. It allows for the collection of various data, including temperature readings of the air before and after contact with animals, temperature of the slurry, and fan rate. Additionally, our experimental station offers services such as measuring individual animal weight, individual carcass composition, and greenhouse gas and ammonia emissions. The ClimatoTec unit has so far only been used in cold conditions and, for instance, has maintained ambient temperatures of $16 \pm 1^\circ\text{C}$ for pigs between 25 and 118 kg. Tests in hotter conditions will also be conducted soon.

From genetic correlation to causation between milk yield, mastitis and health traits in German Holstein cattle

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A comprehensive understanding of the genetic connection between economically important traits in livestock is essential for the sustainable design of breeding schemes. Amongst others, this means going beyond the estimation of genetic correlations and to examine causal relationships, e.g., by performing Mendelian randomisation (MR) analyses. This study aimed to shed more light into the genetic connection between milk yield, mastitis and six claw health traits in German Holstein cattle using a set of 34,497 cows with de-regressed proofs and ~17 million imputed whole genome sequence variants. We performed a bivariate analysis to obtain genetic correlations and a MR analysis to obtain causal associations between milk yield and each of the seven health traits. The results indicate consistency between the correlations and the causal associations regarding the effect directions. They signs were mostly in the undesirable direction, confirming the known antagonism between health and milk production in dairy cattle. However, the estimates differed in the effect sizes and the number of significant results. Significant genetic correlations were found for almost each trait pair, but not all correlated pairings also showed a significant causal association. This highlights the importance of analyses assessing causality between complex traits to improve the understanding of their genetic association and thereby the design of sustainable breeding schemes.

Milk somatic cell transcriptome profile of high and low RFI Holstein cows and the pleiotropic effect on immunity

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Feed accounts for the largest expense on dairy farms, leading to a demand for improving breeding strategies for more feed-efficient animals. As feed efficiency (FE) is a highly polygenic trait, associated genes may have pleiotropic effect on other important traits, such as immunity. This may be a challenge for developing genetic breeding strategies incorporating FE. RNA-Sequencing was used to identify differentially expressed (DE; FDR<0.05, |FC|>2) genes in milk somatic cells of Low-RFI (n=22) and High-RFI (n=17) group of animals from a population of 200 milking Holstein cows. In total, 36 genes were DE between Low- and High-RFI animals. These DE genes were significantly enriched in 28 metabolic pathways (FDR<0.01), the majority of which are associated with host immune response including the RIG-I-like receptor signaling pathway. Using the coordinates of the 36 DE genes, QTL annotation and enrichment analysis was performed and identified 646 previously annotated QTL within the genomic regions of the positional candidate genes. QTL enrichment was also performed to account for the overrepresentation of milk traits in the QTL database. In total, 387 significantly enriched QTL were identified within the gene coordinates, with the majority of the top 10 significantly enriched being associated with health. Results show potential regulatory genes that may impact the variability in FE of Holstein cows, while also having a pleiotropic effect on immunity, which could explain the possible unfavorable moderate genetic correlation between FE and health traits.

Development of a selection index for resilience in German Holstein

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The variance and the autocorrelation of daily milk yield were frequently suggested as indicator traits to improve resilience in dairy cattle. We estimated genetic parameters for four different daily milk yield variance-based indicator traits and for the autocorrelation of consecutive daily milk yields in the German Holsteins. Univariate analysis was conducted for each resilience indicator to derive estimated breeding values (EBV) for each individual. The Pearson method was used to analyze the correlations between EBVs of the resilience indicators and traits implemented in the official breeding value estimation. Subsequently, we combined the five EBVs for resilience indicators with weighting factors in an index, whereby the weighting factors were selected to achieve maximum selection response for health and longevity. The analyses showed that different variance-based resilience indicators contain different information for health and longevity, and that a resilience breeding value should therefore not consist solely of one resilience indicator. An index consisting of five resilience indicators leads to the highest selection response and highest heritability of the index. Two variance-based resilience indicators turned out to be of particular importance for achieving selection response in resilience. In summary, this study revealed that a resilience index can offer additional information and can be a helpful tool for breeders, to select healthy, high and homogeneous performing dairy cattle.

Large scale transmission experiment reveals substantial genetic variation in both host susceptibility and infectivity affecting disease spread and survival

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Genetic selection of animals with low genetic infectivity, in addition to low susceptibility could be an effective method to reduce disease transmission of farm animals. However, to date few genetic parameter estimates for these traits exist, as tools to estimate these traits are still in their infancy. This study combined data from a large-scale transmission experiment in turbot, and novel Bayesian inference software (SIRE 2.1) to provide the first empirical genetic parameter estimates for host infectivity, in addition to susceptibility, as well as infection induced mortality. Validation of SIRE 2.1 with simulated disease and survival data mimicking the experimental design provided reliable (co-)variance estimates and high prediction accuracies for all three traits. Confirming expectations from evolutionary theory, posterior means for genetic variance in host infectivity, estimated from the experimental data, were at least as high as those for susceptibility. In contrast, genetic variation in survival was close to zero. Despite high uncertainties, estimated genetic correlations between the traits were unfavourable. The results highlight the importance for considering multiple host traits affecting disease transmission and survival for genetic control of infectious diseases in farmed animals

Genetic analysis of anogenital distance in dairy cattle

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Fertility is a crucial element of productivity and profitability in the dairy industry, yet genetic improvement of reproductive outcomes has been hindered by low heritability estimates of existing fertility traits. Anogenital Distance (AGD) has emerged as a potential biologically relevant trait for selective breeding programs. Short AGD has been demonstrated to be favorably associated with reproductive outcomes in female cattle. This study aimed to estimate genetic parameters and the reliability of breeding values for AGD. AGD records of 4,988 Canadian Holstein cows and heifers were analyzed using AIREMLF90. A moderate heritability of 0.39 ± 0.04 suggests AGD's potential for selection, surpassing the heritability of current fertility traits. The reliability of the estimated breeding values (EBVs) was moderate to high across analyzed groups of animals (0.51 – 0.83), indicating the possibility for increased response to selection. Further research is warranted to validate these findings and ensure their robustness before widespread implementation. However, our findings underscore the potential of AGD as a heritable trait to enhance fertility outcomes in dairy cattle breeding programs.

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Theatre 6

Analysis of aggressive behaviour in high-production dairy cows through differential expression transcriptome analysis

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Aggression is a deeply ingrained and preserved characteristic common across many animals for purposes such as feeding, self-defence, competition for mating opportunities, and territory protection. The objective of this research was to discover gene expression variations and gene enrichment pathways that influence aggressive behaviour. Additionally, the results were correlated with selection indexes and basic production traits derived from utility assessment and breeding value evaluation of high-production dairy cows using gene whole co-expression network analysis. In the studies, 16 cows on two farms with similar animal husbandry conditions provided biological material. The behavioural phenotype (aggressive vs calm) was determined based on observations. The cortisol level deposited in the hair was determined in the studied group of animals. Subsequently, RNA sequencing was used to identify the expression profile of each cow. The study obtained over 35 million paired-end reads with an alignment average of 90% rate to the ARS-UCD1.3 reference genome. The bioinformatics analysis involved identifying differentially expressed genes in calm and aggressive cows, determining metabolic pathways, and using the GWENA package to determine gene co-expression, characterize individual modules, visualize networks, and correlate animal behaviour with production traits. The analysis showed that a high cortisol level is a characteristic feature of aggressive cows. Based on the analysis of over 9600 identified genes in sequencing, 17 of them were characterized by a high level of differential expression, of which nine protein-coding genes had increased expression. In the co-expression analysis, two large modules of genes characteristic of aggressive and mild cows were identified, containing over 6700 and 1500 genes, respectively. The assigned genes were determined to be characteristic of aggressive behaviours identified in human studies. Pathways included, among others, MAPK and JAK-STAT signalling pathways, as well as pathways initiating immune responses often indicated in considerations related to the occurrence of stress.

Combating psoroptic mange susceptibility in Belgian Blue cattle: can breeding provide a solution?

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Belgian Blue cattle stand out for their remarkable musculature, high-quality lean meat, and efficient feed conversion. However, their susceptibility to psoroptic mange, a skin disease triggered by *Psoroptes ovis* mites, poses significant challenges, leading to skin afflictions that decrease animal welfare and may lead to economic setbacks. The susceptibility to psoroptic mange varies widely among individuals within the breed, suggesting a genetic component to this vulnerability. Through our quantitative genetics study, we shed light on the hereditary nature of susceptibility to psoroptic mange, confirming the long-held belief regarding its genetic basis, and demonstrate that differences between animals cannot be solely attributed to management practices. Moreover, we discuss the feasibility of implementing selection strategies despite the relatively low heritabilities for lesion extent. We critically evaluate the efficacy of the current screening protocols and explore the potential application in practical breeding programs. By emphasizing the novelty of targeting psoroptic mange susceptibility as a welfare trait in Belgian Blue cattle, our study contributes to the broader discourse on enhancing animal welfare through breeding.

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Development of novel traits for genetic evaluation of maternal fertility in extensively farmed beef cattle

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Breeding programmes often include fertility and health related traits alongside more traditional production traits. Traits other than production are, however, often lowly heritable but crucial for sustainable livestock production systems. Reproduction traits have been identified as key economic drivers within New Zealand beef farming systems such that further development and implementation of genetic tools is crucial to assist cattle breeders in making selection decisions. The Informing New Zealand Beef Programme (INZB) is a seven-year programme, jointly funded by Beef + Lamb New Zealand and the Ministry for Primary Industries through their Sustainable Food & Fibre Futures Fund. An important goal of the INZB programme is the development of novel traits, with a key focus on creating a better description of maternal performance and functionality. Fertility-related traits that will be further developed are those with a direct (e.g. conception date) and indirect impact on maternal reproduction (e.g. cow body condition score). Results from the dairy industry have shown that breaking fertility down into its component traits has tended to result in improved heritability estimates and greater scope for genetic improvement. Use of different measurement methods, such as sensor-based livestock monitoring systems, along with common recording practices (i.e. fetal aging) are currently being trialed on two farms to assess their suitability for extensive livestock production systems to predict components of fertility, including age at puberty, conception date, calving date and postpartum anestrus. This work will explore the potential for the further development, practicality, and implementation of these component fertility traits for the genetic improvement of the New Zealand beef population.

Genetic determinism of resilience to unrecorded challenges in pigs

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Selection of animals able to maintain their performance under variable environmental conditions is needed. Pigs in selection farms are raised in a monitored and controlled environment (e.g. temperature). However, pigs could be exposed to challenges/disturbances unrecorded and from unknown origin. Therefore, resilience in pigs remains important. The probability of the occurrence of an unrecorded environmental challenge at a given day was estimated from daily feed intake (DFI) data, and the genetic determinism of resilience to such events was evaluated. In total, 186,242 DFI records recorded automatically from 2,517 Large White males were used. The pedigree involved 5,649 individuals. For each day (within each batch), the probability of the occurrence of a challenge was computed via a mixture model. As a result, unrecorded events of environmental challenges affecting pigs' DFI were identified. Days with a high probability of having a high coefficient of variation (CV) were related to the occurrence of an environmental challenge. The probabilities (p) of high CV (stressful day) were used as a covariate in a reaction norm model (RNM) as an environmental descriptor, ranging from a non-challenging environment ($p=0$) to a challenging one ($p=1$). Variance components were estimated using the average information restricted maximum likelihood method. The mean probability of being a day with a high CV was 0.10; thus most of the days have a low probability of the occurrence of a challenge. The genetic correlation between level and slope was 0.06 ± 0.13 in RNM. This value suggests that selecting for decreased DFI would have a minimal effect on the animal's sensitivity to challenging environmental conditions.

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Comparison of the cytokine profiles after LPS injection between two lines of goats selected for functional longevity

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The present article aims to explore the phenotypic variability of inflammatory reactions and compare them between two lines of goats. Hyper-selection based on the functional longevity of bucks successfully created two groups of goats with different lifespans (High_LGV and Low_LGV). Primiparous goats from the two divergent lines were injected intravenously with LPS (lipopolysaccharide) to stimulate inflammation. Blood was sampled at hours: -72, 0 (injection), 2, 4, 6, 8, 10, 12, 24, 48, 72, and 144. The concentration of 14 cytokines was measured: IFN γ , IL-1 α , IL-1 β , IL-4, IL-6, IL-10, IL-8, IL-17 α , IL36RA, IP10, MIP1 α , MIP1 β , TNF α and VEGFA. The experiment was carried out in two experimental farms (n=83 goats). Results showed that the LPS injection induced a significant increase in several cytokines. The cytokines profiles were different between farms, highlighting different inflammatory backgrounds that could be related to different farm managements. The two longevity lines had different profiles for several cytokines within each farm. Most of the differences between lines were not consistent across farms, showing a potential Gx E interaction. However, the increase of IL-6 was stronger among Low_LGV goats in both farms. These results strengthen our understanding of the immune reactions implied in animal resilience and longevity. This study has received funding from the European Union's Horizon 2020 program SMARTER (agreement No 772787) and APIS-GENE (Paris, France).

Using antibody response to Maedi Visna Virus as selection criterion in sheep

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Breeding programs to increase resistance to Maedi Visna Virus based on recording antibody response have been envisaged. The infectious status (IS) assessed with ELISA test depends on the age at infection and delays in seroconversion. Negative animals early in life may be infected or seroconvert later in their lifetime. This study aimed to evaluate the impact on genetic models of pre-adjusting negative IS recorded early in lifetime. In a naturally infected flock, 3,215 ELISA tests were carried out between 2016 and 2022 on 2,276 genotyped Sarda ewes by 50K BeadChip. The first ELISA test occurred between 9 and 20 mo of age. Ewes with negative IS were re-tested every 6 mo up to culling, while those with positive IS were not. First, the last individual IS (1 positive; 0 negative) was analyzed by a linear animal model including the age class as a fixed effect. In a second analysis, pre-adjustments of negative IS were calculated based on the deviations between the prevalence at 1 or 2 yr of age (45% and 80%) with that at the reference age of more than 3 yr (93%) in a sample of ewes followed up to culling. Pre-adjusted records were weighed for their accuracy based on the mean square deviation between the adjusted and the expected values. Heritability was high for both analyses (0.46) showing the potential effectiveness of selective breeding based on ELISA tests. The pre-adjustment reduced total and genetic variances by 47% confirming that using raw IS records inflates variances. The correlations between GEBV of young ewes with negative records were 0.43 and 0.78 for 1 and 2 years of age respectively, showing that a relevant re-ranking occurs when data are not pre-adjusted.

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Evaluating the influence of selection for disease resistance on the spread of infections in a simulated aquaculture population when underlying epidemiological traits are correlated

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Infectious disease outbreaks are a major threat to the sustainability of the aquaculture industry and thus, selecting for disease resistance is a major objective in aquaculture breeding programs. Disease resistance is usually recorded as mortality in challenge tests performed on sibs of selection candidates. However, it is unknown if direct selection against mortality also reduces disease transmission (i.e., the incidence and severity of outbreaks) given that both mortality and transmission depend on three underlying epidemiological traits: susceptibility, infectivity and infection-induced mortality. This study aimed to evaluate the effect of applying genomic selection against mortality on disease transmission in a simulated fish population. Five scenarios varying in the genetic correlations between the underlying traits were considered. Continuous or binary phenotypes were assumed to be obtained from a sib population challenged with a fast transmission virus. With continuous phenotypes, the test stopped when mortality naturally levels off (with 80% of the fish dead). With binary phenotypes, the test stopped when 50% of the fish were dead. In scenarios with binary phenotypes, disease transmission evaluated as the basic reproductive ratio (R0) greatly depended on the correlation between the underlying traits. In contrast, in all scenarios with continuous phenotypes, selection always led to a reduction in R0 and in most cases reached values < 1. Thus, genomic selection for disease resistance can simultaneously reduce mortality and disease transmission.

Genetic markers associated with bone properties in White Leghorn laying hens

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Bone damage in modern commercial poultry has significant welfare and economic impacts and is recognized as one of the primary challenges faced by the poultry industry. The aim of the present study was to investigate genetic markers that contribute to genetic variance in 44 bone quality and composition traits in White Leghorn laying hens (n=987) using a genome-wide association study. Bone qualities were assessed on live birds and post-mortem using physical methods, X-ray, thermogravimetry (TGA) and infrared spectroscopy (FTIR). We found novel genetic markers (n=30) significantly associated (p-value<10⁻⁶) with humerus breaking strength (chr 20) and density (chr 5), tibia stiffness (chr 1), medullary mineral crystallinity index (chr 1, 10, 13, 27). The annotation of the SNP positions showed their location within proximity to 13 genes, although their function in bone properties is uncharacterized. In addition, SNPs (n=117) with suggestive associations (p-value<10⁻³) were detected as those associated with more than one trait. The overlapped SNPs were detected between postmortem tibia strength parameters (chr: 1, 5, 21, 23, 26). Overlapped SNPs associated with humerus bone strength traits were identified on chromosomes 1 and 20. We identified 26 overlapping SNPs on chromosomes 1, 2, 3, and 6, exhibiting similar effects on tibia strength traits measured in live hens and postmortem. In the case of bone chemical properties, we found 29 SNPs (chr: 1, 2, 3, 4, 7, 10, 11, 21) with overlapped association between medullary and cortical bone compositional parameters measured with both, TGA and FTIR methods. The results highlight genetic markers associated with favorable bone quality traits that can be useful for further functional genomic investigations and breeding value predictions for improving bone strength in laying hens.

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A substitution in the porcine glucocorticoid receptor changes gene expression in the brain and consequently behavior

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Previously, we identified a substitution Ala610Val in the porcine glucocorticoid receptor (GRA610Val) underlying a major QTL for cortisol production in pigs. The gain-of-function of the GR due to the substitution leads to a compensatory downregulation of the activity of the entire hypothalamus-pituitary-adrenocortical (HPA) axis. To gain insight into mechanism of action of GRA610Val and into the regulation of the HPA axis in pigs in general, we analyzed transcriptome changes induced by the substitution in different brain structures under resting conditions, and after application of two different doses of dexamethasone, a selective activator of GR inducing HPA axis suppression. The results show that GRA610Val changes the expression of several clustered protocadherins – cell adhesion proteins playing an important role in neuronal wiring and previously associated with tameness in an animal model of domestication. In addition, the Val variant induces a pro-inflammatory phenotype of microglia – brain immune cells – which is a risk factor for mood disorders. To explore how the expression changes in the brain triggered by GRA610Val influence behaviour, we assessed pigs in standardized tests designed as pre-established indicators of different personality traits (Boldness, Exploration, Activity, Sociability, Coping). We found that pigs homozygous for the Val variant appear bolder and more explorative in a context of novelty compared to the other two genotypes by showing shorter latencies to contact a novel human and novel object, higher latencies to return to home pen and to escape the openfield/novel object test. With regard to the personality trait activity, pigs homozygous for the Val variant also tend to have higher scores as they show higher frequency in leaving and entering the home pen. The changes in gene expression and behaviour induced by GRA610Val have potential implications for welfare and husbandry of pigs.

Genetic analysis of keel bone fractures in laying hens housed in a commercial aviary

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Keel bone fractures (KBF) are a widespread problem in laying hens, with a prevalence of up to 96% in aviary systems. KBF therefore impose a serious threat to animal welfare and the social license of egg production. The causes of KBF are not clear, although age to first egg, total egg production, collisions with housing features, and the shape of the keel are all believed to be related. In addition to environmental factors, it has been suggested that occurrence of KBF has a genetic component. Earlier studies have shown clear differences in prevalence of KBF between distinct genetic lines, and bone traits such as strength and density are moderately heritable. However, genetic variation for KBF within a genetic line has not been studied. Here, we aim to estimate genetic parameters and perform genome-wide association analysis (GWAS) for KBF in laying hens. We will use data on 1,125 white laying hens from a two-way cross of parental lines (Hendrix Genetics). The animals are housed in five groups of 225 animals within a quasi-commercial aviary system with three stacked tiers at different heights and a winter garden. Individual phenotype data include severity of KBF scored as a continuous variable (0.0-10.0) assessed via radiograph images taken at 27 weeks of age. All animals are genotyped with a 60K SNP panel. This study will provide the first estimates of genetic parameters of KBF, and identify putative genomic regions associated with KBF.

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Poster 16

Genetic analyses of blood metabolites and amino acids in blood serum in pigs

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In pig production, resilience is an individual's ability to cope or recover rapidly from a wide range of disruptive challenges back to the original level of normal health, fertility, and production. However, recording resilience in nucleus herds is challenging, as the health status is high, and natural disease challenges rarely occur. To find new methods for measuring resilience on selection candidates, the main goal of this study was to investigate potential bio markers for resilience to disease and investigate potential genetic variation in these markers. Blood samples were collected for 922 pigs from a dam line at the nucleus boar testing station. The samples were taken at entry of the test station, when the animals were weighted, ear tagged and vaccinated. All serum samples were analysed with a targeted analysis, liquid chromatography with tandem mass spectrometry (LC-MS). Univariate genetic analyses were run for each metabolite to identify genetic variation, using linear animal models. Bivariate genetic analyses were run to obtain genetic correlations to growth and a resilience related trait, root mean square error of daily feed intake (RMSE_DFI). Heritabilities ranged from 0.05 to 0.46 for the different amino acids, and the heritability for blood serotonin and kynurenine was 0.28 ± 0.15 and 0.38 ± 0.14 , respectively. No significant genetic correlations were identified between RMSE_DFI and different amino acids, serotonin, or kynurenine. However, a significant correlation between serotonin and growth was identified (-0.7 ± 0.23) and between tyrosine and growth (0.7 ± 0.21). The results suggested that level of specific metabolites and amino acids in blood was heritable, but it was not possible to identify genetic relationships between the metabolites and resilience (measured as RMSE_DFI) in this data set. Further genetic analyses between the metabolites and other health related traits such as osteochondrosis, longevity and exterior will be performed.

Prediction of energy balance of Holsteins in Japan from milk traits including fatty acids

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It is important to improve the energy balance (EB) of dairy cows in lactation to prevent the deterioration of fertility and health. Our objective was to predict EB from milk traits including fatty acids and examine how the fitness of the prediction models varies by lactation period. We used 29 033 records from 92 lactations of 67 Holstein cows for calculation of EB. Milk yield (MY) and feed intake were measured daily, and milk composition and body weight were measured weekly. We defined days in milk class (DIMc) of 7-day intervals from 6 to 306 DIM. Average energy intake and requirement per DIMc were calculated, then EB per DIMc was calculated as the difference between intake and requirement, giving 43 EB values per lactation. We built multiple regression models to predict EB. The independent variables were DIMc, MY, fat, protein, lactose, fat-to-protein ratio (FPR), and 3 types of fatty acid (de novo (DNF), mixed and preformed (PRF)). We created 43 datasets by adding 7 days records each (dataset 1: 6-12 DIM, dataset 2: 6-19 DIM, ..., dataset 43: 6-306 DIM). The optimal variables were selected for each dataset using the stepwise method. The adjusted R² of the model was highest for dataset 1 and over 0.60 for dataset 2-8, then became lower and under 0.40 for dataset 15-43. We selected DIMc, MY, fat, lactose, FPR, DNF and PRF as independent variables in the model for dataset 8 (6-62 DIM). The model for predicting EB using DNF and PRF would be applied to records up to about 60 DIM, since it fits well in the early lactation, but not well including the middle and late lactation.

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Poster 18

Genetic analysis of temperament in Italian Simmental bulls

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Cattle temperament (TMP), defined based on differences in animal's responses to handling or restriction in confined spaces, is directly related to human safety and various economically important traits, such as animal welfare, growth rate, and feed efficiency. The Italian Simmental Breeders Association evaluates TMP in young bulls (1 = docile, 2 = normal, 3 = aggressive). Thus, the aim of this study was to investigate the genetic background of TMP in this breed. The datasets included records from 2,523 young bulls (1,109 genotyped) simultaneously scored by three trained experts. TMP was analyzed as a linear trait because the average values among the three experts resulted in continuous values. The animal model considered the contemporary group as a random effect, twinship and parity of the dam as categorical fixed effects, age at the beginning of the performance test as a linear covariate, and animal as random. The TMP heritability was computed using the gibbsf90+ software; (G)EBV, calculated using both BLUP and ssGBLUP, were validated on the genotyped bulls (344) born in the last three years (2020-2022), for which breeding values were computed considering or not their records. The estimated heritability was 0.10 ± 0.03 . The correlations between (G)EBV estimated considering or not the candidates' records were 0.85 for BLUP and 0.88 for ssGBLUP. Since the bulls of the performance test represent the best males of the population and have complete and verified pedigrees, genomics only slightly increased the prediction accuracy for TMP, which in this breed seems to be under low genetic control as currently evaluated.

Variance components estimation for semen parameters of boars involved in the PDO ham production in Italy
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The dissemination of the artificial insemination (AI) has brought a significant advantage for the animal breeding sector. In particular, the AI has accelerated and spread the genetic progress by increasing the selection intensity, especially for cattle and pigs. Having boars with greater semen doses production capacity allows to further increase the intensity of selection and optimizes the management of AI animals. Thus, aim of this study was to estimate the variance components of semen parameters recorded in Italian Large White, Italian Landrace, and Italian Duroc. The semen has been collected according to a protocol of the Italian pig breeders association (ANAS) and its quality evaluation was determined by the system Htm Ceros-Casa. The following traits were defined for 39,011 ejaculates: volume, concentration, motility, number of doses, morphology (only for 13,024 doses). The phenotypes were analyzed in a multibreed animal model with breed, date of sampling, year of birth as fixed effect, age at sampling as linear covariate, and animal and permanent environment as random effects. All traits had low-to moderate heritability: 0.26±0.03 for volume, 0.14±0.03 for concentration, 0.10±0.02 for motility, 0.14±0.03 for number of doses, and 0.06±0.04 for morphology. As expected, repeatability values were larger, and they ranged from 0.23±0.01 (motility) to 0.31±0.01 (volume). These estimates are in agreement with most of the available literature. Moreover, these low-moderate heritability values are similar to those estimated for other female reproductive traits (e.g., number of born alive, number of farrowing) already included in the ANAS breeding programs.

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Poster 20

Twin birth rate in German Holsteins: related to increased milk yield and fertility level?

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Multiple births in dairy cattle have negative impacts on health and performance of cows and calves. Although the overall prevalence is low, farmers are reporting increased twin birth rate. Twin births are primarily caused by multiple ovulations and, thus, highly correlated to fertility. Moreover, studies have shown that increasing milk yield is associated to higher prevalences of twin births in dairy cows. The aim of this study was to investigate the relationship among twin births, milk yield and fertility at the genetic level in German Holstein cattle. Population wide analyses with over 5.5 million lactating cows showed no increase of twin birth rate over the last decades. The heritability of twin birth was 0.009 (±0.0004), 0.025 (±0.0008) and 0.028 (±0.0009) for calving number 1 to 3, respectively. Initial analyses indicate a low genetic correlation between milk yield and twin births but also low genetic correlations between fertility traits and twin births. To investigate the genetic architecture of twin births, genome-wide association studies were performed, which identified a QTL on chromosome 11 at the LH-CGR/FSHR locus. These genes encode the receptors for the hormones LH, CGR and FSH, which are essential hormones for female reproduction and therefore linked to fertility. Further analyses are required to quantify the impact of the identified QTL.

Genetic perspectives on feeding traits in beef cattle

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Animal feeding and meal patterns significantly impact feed efficiency, which is important for increasing the profitability of beef cattle production systems and can be used as indicators to assess variations in animal performance by breeding programs. This study aimed to estimate genetic parameters for feeding and meal-related traits in Nellore cattle. The data was collected in feeding trials (68 days) conducted between 2013 and 2023. The dataset included information from 1,157 Nellore young bulls with an average age of 283 ± 41 days. The animals were kept in feedlots equipped with electronic feeders that registered the feed behavior of each animal. The following traits were analyzed: dry matter intake (DMI), feeding time (FT), feeding events (FEv), meal time (MT), and meal events (ME). The animal model was used to estimate heritabilities (h^2) and genetic correlations (rg) with genomic information, considering uni and bi-trait analysis. The h^2 for DMI, FT, FEv, MT, and ME were 0.42 ± 0.05 , 0.50 ± 0.05 , 0.39 ± 0.06 , 0.45 ± 0.06 , and 0.27 ± 0.06 , respectively. The genetic correlation were: 0.64 ± 0.07 (DMIxFT), 0.11 ± 0.12 (DMIxFEv), 0.56 ± 0.02 (DMIxMT), 0.24 ± 0.03 (DMIxME), 0.40 ± 0.03 (FTxMT), 0.44 ± 0.03 (FTxME), -0.04 ± 0.03 (FEvxMT), and 0.20 ± 0.03 (FEvxME). Most feeding and meal-related traits were moderately genetically correlated, showing that with more meals per day, young bulls spend more time in feeding activities with higher intake per visit or meal. We conclude that genetic parameters for feeding behavior traits can lead to the genetic selection of animals with more efficient behaviors, resulting in improved feed utilization in Nellore cattle. Funding: FAPESP (#2017/10630-2 and #2023/11176-4) and CAPES (Finance Code 001)

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Poster 22

Antioxidant enzymes mRNA expression in male reproductive tract tissues of European red deer (*Cervus elaphus elaphus*) during rut

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One of the conditions for the proper functioning of the male reproductive system is the balance between production and neutralization of reactive oxygen species (ROS). Previous studies conducted on stallions showed mRNA expression of main antioxidant enzymes (AE) in the tissues of the reproductive system. Knowing that the red deer is a seasonal animal with peak breeding activity lasting only 2 weeks, we assumed that the deer's testicular and epididymal tissues express AE. The RT-PCR was used to analyze expression of SOD 1, 3, CAT, GPx 4, 5 in the reproductive tract tissues. High expression of SOD1, 3 and CAT were detected in cauda epididymis tissues, which indicate the presence of an effective scavenging system that is able to remove excessive ROS production. The highest expression levels ($P < 0.05$) of GPx4 were detected in testis, while GPx 5 expression prevailed ($P < 0.05$) in caput epididymis tissues. The findings of this study provide evidence that the testes and epididymis of the European red deer possess a model antioxidant defense system, which protects spermatozoa against ROS-induced oxidative damage during spermatogenesis, and epididymal maturation and storage. Funded by the Minister of Science under the Regional Initiative of Excellence Program.

Development of a genomic evaluation for teat and udder scores in Angus cattle

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Improving teat size (TS) and udder suspension (US) can have long term benefits for the sustainability of beef cattle herds. Well-structured teats and udders contribute to better calf health and growth, as well as improve cow longevity and milk production. The objectives of this study were to estimate genetic parameters for TS and US, develop pedigree and genomic evaluations, and validate their predictions. Teat size and udder suspension scores were subjectively assessed by breeders following the American Angus Association guidelines. The dataset for variance components estimation included 41,914 scores (for each trait) recorded on 23,886 Angus cows and a total of 154,330 individuals in the pedigree. Using the same phenotypes, the linear regression method was used for validation of pedigree and genomic evaluations including 1.4M animals in the pedigree of which 1.1M were genotyped. The heritability estimates were 0.31 (TS) and 0.34 (US) while the repeatability was 0.51 (TS) and 0.47 (US). The genetic correlation between TS and US was 0.76 showing that the traits are closely related. The prediction accuracy was 0.39 and 0.41 (pedigree), and 0.49 and 0.49 (genomic) for TS and US, respectively. Dispersion was around 0.9, and bias was close to 0 demonstrating no significant over/under dispersion or bias from either model or traits. Considering the amount of data recorded by Angus breeders in the USA, the genetic parameter estimates and the validation results, the implementation of a genomic evaluation for TS and US using a two-trait repeatability model is recommended to help breeders make their selection decisions and improve their cow herds.

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Poster 24

Variance of daily milk yield as an indicator trait in Czech Holstein cows

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Fluctuations in daily milk yield (DMY) could be used as an indicator of the health and resilience of dairy cows. We evaluated the natural logarithm of DMY variance (LnVar) in 2,468 Holstein cows distributed on 9 farms. The data were collected between October 2022 and January 2024. Cows were required to have records for at least 20 successive days with the first day in milk (DIM) <50 and the last day up to 150 DIM. DMY ranged between 3 and 76 kg with a mean of 41 kg of milk per cow and day. The estimation of heritability ($h^2=0.2$) and genomic breeding values (GEBV) was based on a single trait linear animal model and performed with a single-step GBLUP method (BLUPF90 family of programs). The model included fixed effects of the farm*year of calving, the season of calving, the lactation order and the minimum and maximum observed DIM, the random effect of animals (1,878 animals were genotyped with Illumina BovineSNP50 BeadChip) and the random residual. Sires were divided according to their GEBV to lower quartile (Q1, low variance) and upper quartile (Q3, high variance). The data of all their daughters were added from the national database, and their performance in the first three lactations was compared. Q1 daughters had lower milk (-366 kg in first, -142 kg in second and -113 kg in third lactation), protein (-7/-3/-1 kg), fat (-6/0/0 kg) and lactose (-14/-8/-6 kg) production, lower somatic cells count (-10,000/-16,000/-24,000 cells in ml of milk), shorter interval from calving to the first insemination (-1/-1/-1 day) and from calving to conception (-2/-4/-4 days). The two groups did not differ significantly in the number of lactations completed or in the reasons for culling. The study was supported by the Ministry of Agriculture of the Czech Republic, project QK22020280 and institutional support MZE-RO0723-V02.

A genome-wide association study to identify the genetic origin of dermatosparaxis in South African Drakensberger cattle

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Dermatosparaxis is a connective tissue disorder which presents as skin fragility leading to lesions. A causal mutation in Belgian Blue cattle was identified as a deletion in the gene ADAMTS1, while a missense variant in COL5A2 causes the condition in Holstein cattle. The re-emergence of dermatosparaxis in a few herds of Drakensberger cattle, an indigenous Sanga breed, is of concern, with previous studies into the disorder's genetic origin concluding that it is likely a novel mutation. To elucidate the underlying genetic origin of dermatosparaxis in Drakensberger cattle, a case-control GWAS was performed, using 40 genotypes of affected animals in conjunction with the genotypes of over 2000 unaffected Drakensberger animals. All genotypes were sequenced using various commercial Illumina BovineSNP50v3 arrays and a logistical regression association test with permutation were performed. Five SNPs on chr 1 were identified to be significantly associated with dermatosparaxis in Drakensberger cattle. Three SNPs were found to be present in intergenic regions. Of the two remaining SNPs, one was found to be co-located in the TRAT1 gene, which is responsible for the regulation of T-cell receptors, while the second was found to be an intronic variant within SIDT1 which is involved in the immune response pathway linked to psoriasis. Psoriasis is an autoimmune disease which causes chronic skin inflammation in transgenic mice. The results suggests that dermatosparaxis in the SA Drakensberger may have an autoimmune origin.

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Poster 26

A stayability approach for the longevity evaluation of Italian Holstein cattle

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ANAFIBJ (Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana), in the framework of the LATTEco2 project (National Rural Development Programme – PSRN), has been working on updating the genetic evaluation for longevity in the Italian Holstein population. The first step was to define the breeding objective, i.e. the minimum number of lactations a cow should have to have a successful productive life. A stay-ability approach was chosen, coding the stay-ability as a binary trait, where 1 was survived, 0 not survived, for the first four calving events. The full dataset included 9,739,801 cows raised in 37,177 herds and offspring of 110,324 sires. All cows showed a first calving event. The stay-ability curve (rate) was calculated and stay-ability to the fourth calving event was defined as the appropriate breeding goal. A preliminary estimation of breeding values has been performed with a linear animal model: the only random effects were herd-year of first calving, the additive genetic animal and the residual. Bulls' daughters' stay-ability rate (DSR) to the fourth lactation was calculated as a phenotypic indicator. Finally, bulls were divided into three classes: those with EBV higher than 105 showed 58% DSR, while bulls with EBV lower than 95 showed 41% DSR. Further analyses are needed to apply and update the actual longevity breeding value, but the outcomes are promising.

Impact of SNPs for Somatic Cell Count on the Presence of Mastitis Specific Pathogens in Milk

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Mastitis in cattle is often caused by microorganism infections in the udder. Three pathogens causing mastitis are *e. coli*, esculin-positive streptococci (SC+), and coagulase-negative staphylococci (CNS). Samples were collected from 4 farms. Cases and controls were in a ratio of 11:89 for *e. coli*, 20:80 for SC+, and 8:92 for CNS. Cases were reduced due to the presence of other pathogens or multiple records of the same pathogen, resulting in ~1,500 pathogen positive samples. Ten SNPs associated with somatic cell count (SCC) in two previous studies were tested for their effects on pathogen presence (YES/NO). The lasso, backward, and forward methods were used to select SNPs associated to pathogen presence. Two SNPs each were selected for SC+ and CNS, and 3 SNPs for *e. coli*, with one SNP selected for both CNS and *e. coli* (total of 6 SNPs). The concordance index *c* indicated for each pathogen the suitability of the SNPs to separate the samples into cases or controls. The Cochran-Armitage (CAT) and the Jonckheere-Terpstra (JTT) test checked whether a SNP influenced pathogen presence. Finally, a generalized linear mixed model (GLMM) including fixed environmental effects and a random sire effect was fitted to the binary trait of pathogen presence. The CAT and JTT test linked four SNPs to pathogen presence, two of which were confirmed with the GLMM, with effects on CNS and *e. coli*. The joint effects of the SNPs for CNS and *e. coli* explained 13.2% and 13.8%, compared to 19% and 18.4%, respectively, of the full model with all 10 SNPs. Only few SNP genotypes previously linked to lower SCC also decreased the probability for pathogen presence, however, the numbers CASEs and lactations per pathogen were very low in this dataset.

Genome-Wide Association Study on Somatic Cell Count and Health Status of Dairy Sheep

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Somatic cell count (SCC) and mastitis incidence are important traits in sheep that seriously impact the profitability of farms, the longevity of animals, and their welfare. To investigate the genetic background of somatic cell count, a genome-wide association study of selected 40 Lacaune ewes (20 case; 20 control) genotyped with GeneSeek GGP Ovine50k (51873 SNPs) platform was set up. Animals were under intensive farm conditions and official milk recording. Average milk performance of 222,6 kg with 7,31% fat, 5,70% protein and 5,70% lactose were recorded, whereas average SCC was 59174 and 1171483 of unaffected ewes and mastitis ewes, respectively. We identified 20 significant associations with health status and SCC, highest on chr23 (gene SLC14A2) and chr18 (gene CTAGE5). Genes close to detected signals are connected to urea transformation or protein binding functions. Moreover, 13 associations have been observed for milk production with most significant signals on chr1 (genes PTBP2, ROBO1 and PLOD2), chr6 (gene MAD2L1), chr8 (genes PLEKHG1 and PARK2) and chr20 (genes APOM and BAG6) and 1 association for lactose, protein and fat production on chr18 (gene RHCG). Identified genes are included in the genetic control of cell membranes, lipoprotein assembly, ammonium homeostasis or fatty acid synthesis. This work was supported by projects no. 8X23019, DS-FR-22-0016 and APVV-20-0161.

Serological and molecular diagnosis of *Coxiella burnetii* in cows from rural settlements in Brazil

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Coxiella burnetii, widely known bacteria, causes a zoonotic disease, Q fever, and might affect the reproductive capabilities of livestock. The aim of the study was to investigate *C. burnetii* in milk and sera from cows in rural settlements in the state of São Paulo, Brazil. Milk and blood samples from 54 cows in lactation were collected from two rural settlements and tested for molecular analysis and Immunofluorescence Antibody Test (IFAT) for *C. burnetii*. Milk DNA was extracted to perform qPCR, targeting the IS1111 gene, with specific primers IS-202 and IS-249 and probe. All samples were negative at IFAT. At qPCR, nine (16.7%) samples were positive. Results demonstrate circulation of *C. burnetii* in the settlements, showing that DNA bacteria founded in milk have potential to cause Q fever outbreaks, since cattle is an important reservoir for human infection. It is necessary that surveillance be carried out continuously, so rural producers can be made aware and draw the attention of public health agencies to the effects of infection by the agent and contamination of food and environment.

Litter weight gain in pigs has a moderate heritability and favorable genetic correlations to multiple breeding goal traits

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Increased litter sizes in pigs have markedly increased the demand for improving mothering abilities in sows. This study investigated genetic parameters for litter weight gain (LWG) during the first 21 days after litter equalization as a measure of mothering ability in Yorkshire and Landrace sows. We hypothesized that 1) LWG is a heritable trait with a moderate heritability; 2) LWG is favorably genetically correlated to daily gain, feed conversion ratio, conformation, litter size, and maternal effects of piglet growth and piglet survival; 3) LWG is unfavorably genetically correlated to lean meat percentage. Genetic parameters were estimated using restricted maximum likelihood (REML) for one single and two multi-trait models, one including litter traits and one including production traits. We confirmed for the most parts our hypotheses. Thus, we found moderate heritabilities for LWG (0.12-0.19), and favorable correlations to most breeding goal traits. However, exceptions from our hypotheses were noted in the unfavorable genetic correlations between LWG and feed conversion ratio in both breeds, as well as an unexpected favorable genetic correlation between LWG and lean meat percentage in Landrace. These genetic correlations between LWG and feed conversion ratio and lean meat percentage seem to be rather weak or with high standard errors and dependent on breed. In summary, apart from feed conversion ratio, the general pattern was that LWG had favorable correlations with most traits, indicating it could be included into breeding goals without big compromises in genetic gain for other traits.

Direct and maternal genetic analysis for survival at weaning in divergent lines selected for homogeneity in mice

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Selection for homogeneity has resulted in more robust animals resilient to environmental challenges improving the animal welfare. Selection for low variability (L-line) was beneficial for traits usually considered robustness indicators such as birth weight homogeneity, growth, feed efficiency, litter size and longevity. The aim of this study was to study the changes that selection for BW environmental variability can bring to other interesting traits in livestock such as survival rate at weaning (SW). A total of 38,717 SW records from 4,295 litters with 38,718 pedigree records during 34 generations were used. A direct-maternal animal model was used to estimate variance components and to predict breeding values for SW including generation (34 levels), parturition number (2 levels) and litter size (18 levels) as systematic effects, direct and maternal as genetic effect and the litter as random effects. The model was solved by using TM software. Direct and maternal heritabilities were respectively 0.062 and 0.002, with a litter ratio of 0.137 and a non significant direct-maternal genetic correlation of 0.241. The genetic trends were weak and similar between lines for the direct genetic effect. However, for the maternal effect the L-line had about 5 points of higher SW. These results confirmed that reducing the environmental variance of BW also increased survivability at weaning, providing another positive indicator of animal welfare.

Heritability of cumulative mastitis events across productive life in Austrian Fleckvieh cows

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Resilience and resistance to diseases are important breeding objectives to enhance longevity in dairy cows. In this study, we estimated heritability (h^2) and genetic correlation (r_a) of cumulative mastitis events recorded by veterinarians during the lifetime of 18,404 Austrian Fleckvieh cows. Acute (AM) and chronic (CM) mastitis diagnoses were available to count cumulative lifetime events for each cow. As per Austrian legislation, mastitis events refer to clinical forms needing a pharmaceutical treatment officially registered and CM diagnosis can rely on cow's history, e.g., somatic cell count. Genetic parameters were estimated using a linear animal model with 185,948 animals in the pedigree. Cumulative AM and CM ranged from 0 to 8 and 0 to 7 and their h^2 was 0.09 ± 0.01 and 0.01 ± 0.00 . The r_a with milk yield was -0.10 ± 0.05 for AM and -0.34 ± 0.10 for CM. Results suggest that susceptibility to mastitis is heritable and that cumulative AM may be an additional proxy to drive selection towards more resistant animals.

Embryo production efficiency in divergent lines selected for homogeneity in mice

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A divergent selection experiment for birth weight environmental variability has been successfully conducted during 34 generations in mice. Animals from low variability line (L-line) have been shown to be more robust in the sense of having higher litter size, survival, growth, feed efficiency and longevity in a common breeding environment, than those from high variability line (H-line). Differential litter size in the lines can be due to embryo production, embryo survivability or both. The objective of this work was to analyse the differences between these divergently selected lines regarding the number of embryos produced. A total of 15 males per line and 128 females (59 for H-line and 69 for L-line) were used, each male with 3, 4 or 5 full-sib females designing a minimum coancestry mating inside line (Endog 4.9), in order to perform in vitro fertilization and cryopreserve the embryos. A linear model was fitted to analyse the total number of embryos obtained, including the line, the number of females per male and the female age as effects. The Pearson correlations between the traits were also analysed. There were significant differences for the number of females and the female age ($p < 0.05$), but the line effect did not affect the number of embryos; the correlation between the number of embryos and the female age was 0.65 ($p < 0.001$). These results were consistent with previous reports in this experimental population that found higher ovulation rate in H-line, no difference in number of embryos at 14 days of gestation, and higher litter size in L-line, showing higher embryo and foetal survival in L-line.

BERV-K1 like element and viral infection in Italian Mediterranean River Buffalo: preliminary results

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BERV-K1 (Bovine Endogenous Retrovirus K1) is a proviral element containing full-length envelope (env) belonging to the genus Betaretrovirus and probably integrated between 18.3 and 25.4 million years ago in the genome of Cattle. The integration of viral genome fragments in the nuclear genome of a host (Endogenous Viral Elements – EVE) is a well-documented phenomenon that can provide functions as conferring immunity or increasing susceptibility to viral infection. A BERV-K1 like element with identity >90% on *B. bubalis* chromosome 9 has been previously identified. To test the effect of this EVE on the susceptibility to viral agents of Mediterranean Italian River Buffalo, the number of copies of the EVE were assessed through qPCR performed on 50 buffaloes that were also tested for positivity to BuHV-1 (Bubaline herpesvirus 1), BuCov (Buffalo Coronavirus) and Reovirus (Blue Tongue Disease). Our preliminary results showed some significant association with BuCov and Reovirus infections. Acknowledgements: Research funded by the Ministry of Health – General Directorate of Animal Health and Veterinary Drugs. prog. IZS ME 03/20 RC

TNF α expression at mRNA and protein levels in quarters adjacent to the infected with staphylococci in dairy cattle udder parenchyma

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Cytokines modulate immunity and mediate many functions in organisms. TNF α regulates some biological processes (cell proliferation, differentiation, coagulation, apoptosis, lipid metabolism, induces other cytokine production). The study aims to analyse TNF α expression (mRNA expressed in the ln scale, protein conc. pg/mL) in parenchyma of udder quarters adjacent to infected with coagulase-positive (CoPS) or coagulase-negative (CoNS) staphylococci, compared to fully healthy udders. Cows culled at the end of lactation had either reproductive problems without mastitis or chronic recurrent and incurable subclinical mastitis. Of 200 samples, 10 were from quarters adjacent to CoPS (AHCops), 10 from CoPS infected, 10 from adjacent to CoNS (AHCNS) and 10 from CoNS infected. Ten samples from the healthy udders served as controls (H). The RT-qPCR and ELISA methods were used to establish TNF α expression. For statistical analysis, a MIXED procedure (SAS) was used. The model involved a random cow effect and group and parity fixed effects. We found lower mRNA and protein levels in AHCops than in CoPS while lower mRNA in H than in CoPS with similar protein levels in both groups. The higher mRNA levels in AHCNS than in H and AHCops were stated, while protein level was higher in AHCNS than in CoNS, H, and AHCops. We suppose that CoPS infection lowers and CoNS increases TNF α production in adjacent quarters. Despite the low mRNA level in H, the protein concentration did not differ from CoPS and CoNS. Protein levels in AHCops did not differ from H. It might mean that CoPS infection has no impact on the TNF α level in AHCops, whilst TNF α expression in AHCNS was higher than in H. It may mean that CoNS impacts TNF α expression in adjacent quarters. Financed by NSC, Poland; project No. 2020/39/O/NZ9/02519

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Poster 36

Indicators for energy balance “hidden” in historic data to improve current breeding models in cows

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Fertility deficiencies in high yielding cows are a major reason for farmers to restock their herds. Breeding companies have addressed this issue by providing breeding values for health and fertility trait complexes. However, the phenotypes used focus on easy-to-measure characteristics, whereas the underlying biology is usually not considered. Biology-wise it is known that the energy balance (EB) of the cow is a key for successful insemination. Including indicators for EB in the current breeding goals would therefore potentially improve the respective trait complexes. Work on this has been done by using individual dry matter intake data, which is still expensive for large-scale recording. In the Netherlands a large amount of historic data is available, including individual daily body weights, MIR profiles, genotypes, milk performance, and other data from automated milking systems. This data has been recorded over decades, across parities and lactations stages, is easy and cheap in ongoing recording, but has not been explored to its full potential yet. On this note, a novel phenotype for EB will be developed to combine these data sources for subsequent use as an auxiliary trait in the breeding value estimation. By use of a multi-trait model between this EB phenotype and EB related traits like health and fertility, prediction models will potentially indirectly benefit. This work was supported by the Dutch Ministry of Economic Affairs (TKI Agri & Food project LWV20054) and the Breed4Food partners CRV, Hendrix Genetics and Topigs Norsvin.

Genetics of twinning rate in Italian Holstein cattle

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Twin birth in cattle affects calving ease and metabolic aspects of the dam and is a risk factor for abortions and survival of calves, leading to non-negligible economic losses. To determine the potential role of selection against twinning, we estimated variance components for twinning rate in Italian Holstein breed under a Bayesian framework using 1.2 million calving events in 1,830 herds. Calving phenotype was treated as a binary trait (0 = single birth; 1 = twin/triplet birth). Linear and threshold animal models were used to analyze calving phenotype with parity as fixed effect, and herd-year-season, permanent environmental, additive genetic, and residual as random effects. A multiple-trait approach was used to estimate genetic correlations across different parities (1, 2, 3, ≥ 4). Iteration until 150,000 rounds was performed with GIBBSF90+ and post-Gibbs analysis was conducted through POSTGIBBSF90 (burn-in: 50,000 samples, thinning interval: 200 samples). The twinning rate in the population was 2.7%. Heritability was 0.017 ± 0.001 and 0.063 ± 0.006 , and repeatability 0.018 ± 0.001 and 0.068 ± 0.006 for the linear and threshold approach, respectively. Genetic correlations were >0.80 , with the strongest association between parity 2 and ≥ 4 (0.96). The non-zero heritability suggests a basis for genetic selection against twinning in Italian Holstein cattle.

Session 85

Poster 38

Genetics of Body Condition Score in five Italian beef cattle

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Genetic improvement in beef cattle has been focused mostly on productive trait; however, reproductive and functional traits (body condition, locomotion, maternal ability etc.) have gained interest in recent years, particularly in beef cattle in open pasture and loose system. In this context, body condition score (BCS) is an indicator of body energy reserves after grazing period and it is an indicator of grazing aptitude. This study was aimed to estimate genetic parameters for BCS traits in Marchigiana (M), Chianina (C), Romagnola (R), Maremmana (MM) and Podolica (P) Italian beef cattle. BCS was identified in 4 traits: BCS total (BCSt), BCS ribs (BCSri), BCS loins (BCSI), BCS rump (BCSru), through linear type traits evaluated on cows using a 1-5 scale system, i.e., from 1, very thin, to 5, very fat. Data obtained from the ANABIC database included 25490 M cows, 22445 C, 4796 R, 5622 MM, and 16193 P. A preliminary GLM allowed the identification of major factors affecting the scores, identified in the herd-year classifier, the age at evaluation (in classes), the distance from calving, and the season of evaluation (4 levels). BCSt heritability (h^2) varied from 0.04 to 0.09, whereas h^2 for BCSri varied from 0.03 to 0.07. BCSI h^2 ranged from 0.02 to 0.09, while BCSru h^2 from 0.03 to 0.08. Medium/high genetic correlations among the 4 BCS traits were found in all breeds by multiple traits analysis ranging from 0.61 to 0.98. This study suggests a low-moderate genetic component of BCS in these breeds, but still interesting to be used in breeding for functional traits. Funding: Progetto I-BEEF2 — PSRN Sottomisura 10.2 2014-2020.

Genetics of milkability obtained from milking machine in the local dual-purpose Rendena cows

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Milkability (MAB) is the rate at which milk is fully drawn from a cow's udder, and it is an important functional trait directly related to milking cost and udder health. This study aimed to analyse the genetic components of MAB and its genetic correlations with functional traits such as somatic cell score (SCS), fertility (Days Open; DO), and functional longevity (LONG) in Rendena cattle—a small local dual-purpose cattle breed of the Italian Eastern Alps. Records of MAB were collected directly from milking machines between December 2022 and November 2023, involving 453 Rendena cows across 12 farms, with a total of 49,435 records analysed. MAB showed a moderate-low heritability (0.11 ± 0.07). Genetic correlations with SCS, DO, and LONG were low to moderate, although unfavourable, with values of 0.09, 0.22, and -0.09, respectively. In practical terms, the results confirm that an increase in MAB (i.e., milk flow rate) is linked with a small but unfavourable decrease in udder health, fertility, and functional longevity, even in a local cattle population like Rendena.

Correlations between early-in-life fertility traits and linear type traits in Holstein-Friesian dairy cattle

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Age at puberty (AGEP) and anogenital distance (AGD) are moderately heritable traits, which may have value as early genetic predictors of fertility in cattle. It is important that selection on younger AGEP and shorter female AGD do not negatively affect other traits. We categorised AGEP and AGD and their genetic and phenotypic relationships with a range of body and udder conformation traits. Age at puberty and AGD were measured in 5,010 Holstein-Friesian (HF) and HF × Jersey (J) crossbred yearling heifers from 54 pasture-based dairy herds in New Zealand. During first lactation, a subset of animals from 17 herds were scored for 18 linear type traits ($n=1,403$) and their AGD measured ($n=1,956$). Genetic correlations between AGD and udder conformation traits were weak (-0.10 to 0.13), whereas those between AGEP and most udder traits were moderate and positive (0.18 to 0.46). Except for rear leg set (-0.37), genetic correlations between AGD and body traits were generally weak (-0.21 to 0.23) and not different to zero. Capacity traits had genetic correlations with AGEP that although weak, differed from zero (-0.26 to -0.20) and stature had a positive genetic correlation with AGEP (0.44). Phenotypic correlations for AGEP and AGD with all body and udder conformation traits were near zero (-0.07 to 0.09). These results suggest that selection on younger AGEP or shorter AGD in HF and HF×J heifers should not have negative consequences for body conformation, but AGEP may have small negative consequences for udder conformation. Further work on other important traits and in other breeds should be undertaken to determine the suitability of these early-in-life fertility predictors in breeding programmes.

Effects of low doses of antibiotics on the chicken transcriptome

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The concern related to the use of antibiotics in food-producing animals is significant, with antimicrobial resistance posing a growing threat to public health worldwide. One of the primary problems associated with the overuse of antibiotics is antimicrobial resistance (AMR). The diverse range of bacterial species in the gut microbiota, each with varying levels of susceptibility, means that even low doses of antibiotics administered through cross-contaminated feed can select for AMR. This selective pressure can lead to the development of antibiotic-resistant bacteria, which poses a threat not only to animal health but also to human health through the food chain. Therefore, a study was undertaken to verify the potential impact of low antibiotics given to broiler chickens during the whole rearing period on the gut health identified by the transcriptome of selected immune and metabolic-related organs. Six different antibiotics: colistin, doxycycline, flumequine, thiamphenicol, tiamulin, and tilmosin were administered in the feed at doses equal to 1% of the maximum approved concentration and compared to the control group. The impact of the low doses of antibiotics on the host organism was analyzed based on the gene expression signatures of mucosal scrapings (from cecum), liver, spleen, and pancreas. (This project is funded by the National Research Center UMO-2021/41/B/NZ9/04114).

Identification of functional SNPs causing alternative splicing events linked to subclinical intramammary infection in Holstein cattle

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Functional variations such as SNPs and INDELs in transcribed regions can impact alternative splicing leading to different mRNA isoforms. Aim of this study was to identify potential functional variations causing those alternative splicing events which may explain differences in cattle mastitis susceptibility. RNA-Sequencing was performed of milk somatic cells samples collected from healthy Holstein cows negative to bacteriological test (n=9; NEG) and with natural subclinical intramammary infection due to *Streptococcus agalactiae* (n=11; Sa+) or *Prototheca* spp. (n=11; P+). Functional genetic variants were identified using the ARS-UCD1.3 bovine reference genome following an optimized RNA-Seq variant calling pipeline using STAR aligner and BCFtools for the variant calling. Preliminary results revealed several SNPs uniquely fixed in the Neg group, Sa+ and P+ group. Also, a more in deep analysis to identify the SNPs that are present in extreme frequencies across the groups are under study. In addition, to SNPs, further analyses will focus on the detection of potential INDELs causing those alternative splicing events linked to subclinical intramammary infection in Holstein cows. Functional analysis including the identification of splice sites events and the associated functional consequences will be performed using CLC Bio Genomics workbench (QIAGEN) and Variant Effect Predictor (Ensembl). With enhanced functional genomic insights into the regulation of subclinical intramammary infection this could optimize selection in dairy cattle industry to improve overall bovine health and productivity. Acknowledgements: Agritech National Research Center, funded by the European Union Next-Generation EU (Piano Nazionale di Ripresa e Resilienza (PNRR)–Missione 4, Componente 2, Investimento 1.4–D.D.1032 17/06/2022, CN00000022)

Genetic analysis of predicted negative energy balance and its biomarkers of first-parity Holstein cows in early lactation

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Negative energy balance (NEB) state has negative impacts on milk production and health in dairy cows. Although direct detection of NEB in dairy cattle is very expensive, researchers have identified multiple biomarkers to detect NEB. However, the genetic relationship between NEB and these multiple biomarkers remains unclear. The aims of this study were therefore to (1) estimate variance components for predicted NEB (PNEB), the Cluster4 (highly related to NEB, it is an index based on multivariate analysis of milk mid-infrared predicted biomarkers), 15 biomarkers related to NEB, and 3 production traits (milk yield, fat percentage, and protein contents) in first-parity Holstein cows in the early lactation (DIM from 5 to 50 d); (2) calculate the genetic correlations among them; (3) estimate the causal effects of PNEB on these 19 traits through a recursive model. After quality control, the predicted dataset comprises 30,364 records on 25,287 first-parity cows. The PNEB and 15 of 18 studied traits were predicted using milk mid-infrared spectral. Results showed a moderate h^2 (range 0.16 to 0.29) for all traits, except for predicted citrate (0.38). The genetic correlations between PNEB and Cluster4, 15 biomarkers, 3 production traits range from -0.60 [predicted blood insulin-like growth factor 1 (IGF-1)] to 0.87 [predicted blood non-esterified fatty acids (NEFA)]. The PNEB showed the highest causal impact on NEFA. Nine (Cluster4, blood NEFA, blood Beta-hydroxybutyrate, blood insulin-like growth factor-1, Acetone, C18:1cis9, Long chain fatty acid, Citrate, Milk yield) traits have been suggested as highly relative to PNEB. In further development, through the studied relationships, the study will help us better understand the genetic mechanism of NEB.

Effect of E35K TMEM154 variants on the genetic variability of antibody response to Maedi Visna Virus in Sarda sheep

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The E35K polymorphism at TMEM154 gene has been associated with genetic resistance to Maedi Visna Virus (MVV) in sheep. A significant polygenic variation ($h^2=0.46$) of antibody response to MVV was previously assessed in the Sarda breed. This study aimed to evaluate the association of E35K polymorphism with genetic resistance to MVV and its impact on the total genetic variability. In a naturally infected flock, 3,215 ELISA tests were carried out between 2016 and 2022 for assessing the infectious status of 2,276 ewes genotyped for E35K TMEM154 alleles and by 50K BeadChip. The first test occurred between 9 and 20 mo. Then, negative IS ewes were re-tested every 6 mo up to culling, while those positive were not. Frequencies were 89% and 11% for E and K alleles. An unfavorable dominant effect on IS of E allele was observed with a prevalence of 1523/1864, 246/306 and 4/106 for EE, EK and KK genotypes. Last IS (0: negative; 1: positive), pre-adjusted for the age at sampling, was analyzed with a linear animal model including the genomic relationship matrix and E35K genotype class as fixed effect (EE and EK vs KK). Heritability was 0.34, 26% lower than that neglecting the E35K genotype effect. Results confirmed a strong effect of E35K polymorphism genotype on the antibody response to MVV (EE and EK: 0.93; KK: 0.23). Selective breeding based only on E35K genotype may be challenging due to the low frequency of K allele and the dominant effect of E allele. Therefore, considering the relevant polygenic variability even when accounting for the E35K genotype effect, combining polygenic breeding values with E35K genotype information may be an option to increase genetic resistance to MVV.

Genetic analysis of longevity in three sheep breeds

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Sheep are highly productive animals, yielding valuable commercial products such as meat, milk, and wool. Enhancing the longevity of sheep is crucial for reducing overall livestock maintenance costs and maximizing profitability. Understanding the factors influencing ewe longevity involves analyzing the sequence of survival events from birth to maturity and subsequent parity events. In this study, we estimated the variance components of survival in the first four parities for three sheep breeds: Merino, Heidschnucke (moorland sheep from northern Germany), and German Whiteheaded Mutton. The dataset comprised 56,901, 15,573, and 6,874 observations for Merino, Heidschnucke, and Whiteheaded Mutton, respectively. The pedigrees consisted of 65,387, 18,491, and 9,234 for Merino, Heidschnucke, and Whiteheaded Mutton, respectively. Variance component estimates were obtained based on bivariate animal models using Wombat software. Observations were treated as binary, whenever a given ewe achieved a successful parity the observation was marked as 1 and 0 otherwise. Herd-year, age and season were the fixed effects in the model, and animal additive genetic effect was random. For Merino, the heritability estimates ranged between 0.024 to 0.029, and the additive correlations ranged between 0.5 to 0.8. The heritability estimates for Heidschnucke ranged between 0.008 to 0.028 with additive correlations between -0.08 to 0.8. For Whiteheaded Mutton, the heritability estimates ranged between 0.008 to 0.14 with additive correlations between 0.7 to 0.9. The results indicate that a proportion of the variation in longevity in these three sheep breeds could be attributed to additive genetic effects and thus it is feasible to improve this trait through genetic selection. However, further analysis is recommended.

Session 85

Poster 46

A multi-breed genome-wide association study for temperament in three Italian beef cattle breeds

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Marchigiana, Chianina, and Romagnola are local Italian beef cattle breeds characterized by large dimensions, having the real possibility of hurting personnel and properties during management. The aim of this work was to detect potential genomic associations for the temperament phenotype by using 150K chip genotyping data of young bulls in performance test at ANABIC genetic station. Temperament was recorded as a linear score during 12 different moments and manipulations by assessors, and the final values were rank-based transformed. After quality control, a multi-breed dataset of 228 animals (84 MAR, 78 CHI, 63 ROM) and 117,662 SNPs remained for genome-wide association study (GWAS). GWAS was performed with GEMMA software, using a univariate linear mixed model, considering breed code, year, and month of birth as covariates. The results highlighted a significant SNP (rs137113406) on BTA23 to be associated with temperament (P-value 1.30E-7). This SNP mapped inside a cluster of olfactory receptor genes, which are known to be related to animal behaviour and responses to stimuli. Further studies will be necessary to validate this locus and estimate other QTLs associated to docility. Selection on temperament traits could improve animal welfare, social sustainability, and animal productions. Funding: Italian Biodiversity Environment Efficiency Fitness— I-BEEF 2 — PSRN 2014-2020

Session 86

Theatre 1

Advances in improving chicken welfare from hatch to slaughter

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As a society, we increasingly prioritise the welfare of animals kept for food production. When assessing animal welfare and implementing welfare improving strategies, we need to consider the entire lifespan of the animals, in case of chickens from hatch to slaughter. In this presentation I share the results of various current studies of my group on this topic. Conventional hatching in hatcheries without access to feed and water can affect broiler chick welfare. Therefore, alternative strategies have been developed, for instance providing chicks with early nutrition in the hatchery or hatching eggs directly on-farm. Our results indicate that conventionally hatched chicks scored significantly worse for footpad dermatitis later in life than chicks hatched in alternative systems. However, the hatching system had minor effects on other welfare indicators, such as fear responses in behavioural tests. For the period of life that chickens spend on the farms, we have developed various sensor and vision-based methods to automatically and continuously measure changes in activity and gait as welfare indicators. By determining automatic gait score distributions we have achieved results that better reflect the status of the individual animal in a group than a simple average gait score of a flock. For laying hens, we are currently developing a vision-based approach for the automated tracking of individuals. The results will be used to implement strategies that reduce harmful behaviours like feather pecking and promote positive social interactions. Another project that focusses on assessing animal welfare by automated monitoring and feedback at different stages of the production chain. Here, we link data collected on farms to indicators automatically measured at the slaughterhouse, such as footpad dermatitis and wing fractures.

Session 86

Theatre 2

Perinatal management and dietary strategies improving embryonic and post-hatch development of broilers

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Broiler chicks, during their early life, require careful attention to ensure optimal development so they can reach an optimal performance. Various strategies have emerged to provide these chicks with a strong start. As the gut continues to mature after hatching, and as it becomes colonized, the chick's immune system—initially relying on passive immunity—undergoes further development in the post-hatch period. More and more, the focus lies on assisting chicks during this critical perinatal phase, with the ultimate goals of promoting gut health, minimizing infection risks, and achieving uniform flock growth. In ovo injections of essential nutrients and bioactive molecules are actively researched. These injections aid in gut maturation and colonization, setting the stage for healthy digestion, nutrient utilization, or aid in energy supply and contribute to enhanced muscle development. Furthermore, strategies that focus on the hatching window can significantly impact chick health. Ensuring early access to feed and water immediately after hatching is essential. This promotes robust growth and vitality. Probiotics, whether administered in ovo or shortly after hatching, offer another a powerful approach. They facilitate communication between early microbial colonization and the development of innate immunity within the intestinal tract. Interventions during the perinatal period will further contribute to broiler chick success. By implementing these diverse strategies, we can optimize growth, reduce health risks, and contribute to a thriving broiler flock.

Study of inter- and transgenerational effects of in ovo nutriepigenetic stimulation on chicken cecal mucosa transcriptome

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The research aimed to investigate inter- and transgenerational effects of in ovo embryo stimulation on Green-legged Partridge-like chicken cecal mucosa transcriptome. Synbiotic PoultryStar® (Biomim) and choline were injected in ovo on the 12th day of egg incubation. Three groups were established in generation F1–1/control (C); 2/synbiotic (S); 3/synbiotic combined with choline (SCH). In the generations F2 and F3 groups S and SCH were divided into two groups, each–A/single injections in F1; B/repeated injection scheme in each successive generation. Cecal mucosa samples were collected from 39 birds at 21 weeks post-hatching. RNA-seq libraries were sequenced with an Illumina NovaSeq 6000 platform (PE150). Paired-end reads were mapped to the chicken reference genome (*Gallus gallus* bGalGal1.mat. broiler.GRCg7b). Differential expression analysis was conducted using DESeq2 v.1.42.0. In general, in ovo injections of bioactive substances resulted in changes in the expression of specific genes in the cecal mucosa of adult birds. Transcriptomic analysis revealed differences in genes related to the cellular lipid catabolic process in generation F1, vascular smooth muscle contraction, and the cell surface in generation F3 in response to the synbiotic injection. Combined synbiotic and choline administration modulated expression of genes related to chromosome condensation, lytic vacuole, lysosome, cellular response to stimulus, circulatory system development pathways in generation F2, brush border membrane, and organic acid transport in generation F3. Especially in F2, DEGs were pronounced in the cecal mucosa of groups injected with synbiotic combined with choline. Further, deepened analysis will explore how the synbiotic and choline stimulation in ovo may regulate processes in the cecal mucosa through the indicated pathways. This research was supported by the National Science Centre, Poland (grant no. 2020/37/B/NZ9/00497).

Session 86

Theatre 4

Changes in gene expression and DNA methylation in the liver of broiler chickens after in ovo stimulation with sodium butyrate

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The liver is an important organ that performs immunological and metabolic functions in the organism. The liver and the intestines form a bidirectional relationship known as the gut-liver axis. The intestines and liver communicate via the portal vein and the systemic circulation. Metabolites produced in the intestines are transported via the portal vein to the liver. At the same time, the liver transports bile salts and antimicrobial molecules into the intestinal lumen via the bile ducts. Butyrate is one of the three main short-chain fatty acids, and its functions include providing energy, controlling the state of the intestinal microbiota and mediating the immune response. External sodium butyrate supplementation improves poultry production parameters and stimulates dynamic changes in the intestinal microbiota, which can, directly and indirectly, affect the liver. The aim of the study was to analyze the gene expression and methylation in the liver after in ovo stimulation by sodium butyrate on day 12 of egg incubation in broiler chicken. Incubated eggs of Ross 308 broiler chickens on day 12 of incubation were injected with saline as the control group and with postbiotic – sodium butyrate (SB; three doses: 0.1%, 0.3%, 0.5%) in the experimental groups. The liver was collected postmortem on day 42 after hatching for RNA and DNA extraction. Gene expression analysis was performed by RT-qPCR, while gene methylation by MS-qPCR for a panel of genes: ANGPTL4, CYR61, KLHL6, SYK, and NR4A3. Stimulation with bioactive substances caused changes in the gene expression level, including an increase in the ANGPTL4 and NR4A3 genes in the 0.1% and 0.3% SB groups and down-regulation of the ofCYR61 gene in the 0.5% SB group. In the case of DNA methylation, its level is usually correlated with the level of gene expression. As the methylation level of a gene increases, its methylation decreases. Sodium butyrate affects changes in the level of gene expression and methylation in the liver, which may indicate its epigenetic potential. The study was financed by grant UMO-2021/43/D/NZ9/01548 financed by the National Science Center in Cracow (Poland).

Influence of Hypoxia During Incubation on Broiler Embryo Metabolism and Development: A Tale of Two Genetic Lines

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The modern broiler is raised to maximize its genetic potential, promoting rapid growth. However, enhancements in performance have been linked to increased tissue development at the expense of critical physiological functions like thermoregulation. Consequently, modern broilers face challenges in effectively managing energy expenditure. To assess the impact hypoxia on modern broiler embryo metabolism and thermal ability two genetic lines were used: modern broiler line and a distinct genetic line that has not undergone selection for nearly four decades (since 1986). At embryonic day 16 (E16), broiler embryos from these two genetic lines were subjected to two treatments: 1. control; 2. 17% O₂ concentration for 12 hours daily from E16 through E18. Comparative analysis revealed significant differences between the two lines. Monitoring eggshell temperature and heart rate as physiological indicators revealed a notable increase in response after 2 hours of hypoxia. By the final day of hypoxia (E18), a modest or non-significant rise in heart rate was observed in both lines embryos, indicating adaptation to low-oxygen conditions. Additionally, an increase in hematocrit values was noted, with the modern broiler embryos displaying milder reaction. Both heart rate and hematocrit responses suggest greater plasticity in the 1986 line embryos, which exhibited a more pronounced response compared to the modern line embryos. Examination of organ development revealed that modern broiler embryos maintained their breast muscle integrity throughout the stress period, whereas the 1986 line embryos postponed breast development to support other tissues. The 1986 embryos were able to achieve higher oxygen consumption, demonstrating their ability to cope with the stress, by providing tissues more oxygen in order to function. Genes expression Patterns in the embryo's heart revealed that the modern broiler embryos must increase energy production, at a much earlier stage comparing the embryos of 1986 Hypoxic exposure during incubation initiate a cascade of physiological responses that led to adaptive metabolic plasticity' that may improve modern broiler ability to allocate energy, particularly under suboptimal ambient temperatures.

Session 86

Theatre 6

Classification, Evaluation, and Control of Myopathies in the Broiler Chicken

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Artificial selection for fast-growing and high-breast-yield hybrids has considerably marked up the pressure on breast muscle development, leading to the appearance and expansion of growth-related breast meat abnormalities myopathies (i.e. White Striping, Wooden Breast and Spaghetti Meat) affecting the pectoral muscle of heavy and fast-growing birds. Occurrence of these muscular abnormalities negatively impact both visual aspect and quality properties of raw and processed meat, causing relevant economic damages for the poultry industry. In the past few years, several studies have been carried out to investigate the biological and genetic mechanisms involved in their occurrence. Main features deal with hypoxia, oxidative stress, dysregulation of energy and carbohydrate metabolism, metabolic shift, vascular damage, and muscle development. It is commonly recognized that the occurrence of growth-related abnormalities boosts with increasing growth rate, slaughter age as well breast yield and weight. Within this context, it seems that artificial selection for broilers growth is close to biological limits and further improvements might be restrained by muscle biological potential and related animal welfare concerns. Thus, attempts have been made in the field of animal nutrition to reduce the occurrence of abnormalities through the modulation of both feed formulation (i.e. dietary supplementation of antioxidants, organic minerals, vitamins and aminoacids) or dietary intake through feed restriction. In this scenario, it has been also recently suggested that particular attention should be given on the modulation of embryonic formation of additional myofibers, instead of relying on post-hatch selection aimed at increasing muscle mass accretion.

Multicriterion assessment of two scenarios designed to improve animal welfare in French conventional broiler production

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In broiler production, solutions to improve animal welfare (AW) are currently proposed and discussed in response to societal demands and scientific advances. Yet, implementing new equipment or practices in farms can have positive and/or negative effects on sustainability issues other than AW. To assess the possible trade-offs in scenarios designed to improve AW, we developed a multicriterion method with various French stakeholders (operators, farmers, NGOs, R&D). The assessment grid was structured around four pillars: AW, Economy, Environment, and Farmer. Each pillar was divided into 2-3 components, which in turn were described with criteria (total of 23), each one being measured by a single indicator. With this method, two scenarios of evolution of French conventional broiler production were tested and compared to a “reference” one (Ref.). In the first scenario (Conv+), environmental enrichments (pecking blocks, platforms) were added in the broiler house compared to Ref. In the second one (ECC), in addition to these enrichments, animal density was decreased down to 30 kg/m² (vs. 39 kg/m² in Ref.) and a slower-growing strain was used (Redbro vs. Ross 308 in Ref.). Negative effects were limited in Conv+, while economic and environmental indicators were greatly impaired in ECC, despite a clear improvement in AW. In Conv+, enrichments increase annual working time, but not in ECC because of a lower annual number of flocks. The various levers combined in ECC limit work arduousness and both scenarios slightly improve the farmer’s satisfaction with his/her production system. These results will help stakeholders to make strategic choices, by establishing the best compromises according to their own objectives.

Session 86

Theatre 8

ProBioHuhn: Variations in the gut microbiome of three organic chicken types across different fattening periods and environments

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While knowledge of organic chicken farming continues to grow, there is still limited understanding of the variation in the gut microbiome of organic chickens. This study aims to elucidate the gut microbiome differences across organic farms, examining three fattening types (slow-growing broilers, layer males and dual-purpose males) and multiple time points within the fattening period. Samples from meconium and cloaca swabs were collected and analysed with target amplicon sequencing. Shannon diversity index revealed lower diversity in meconium than in cloaca samples. Principal coordinate analysis identified distinct clustering of meconium samples away from cloacal samples, with the latter showing age-related diversity, suggesting age as a key factor in microbiome composition variation. Microbial composition at the genus level differed significantly between sample types. In meconium, *Enterococcus* (64%), *Escherichia-Shigella* (13%) and *Clostridium sensu stricto 1* (11%) were predominant, while cloacal samples were dominated by *Lactobacillus* (19%), *Ligilactobacillus* (11%) and *Enterococcus* (11%). This study provides an overview of the chicken microbiome in organic farming, highlighting the influence of the development stage and fattening type on microbial diversity and composition.

Synchronizing feed availability with the natural rhythm improves productivity and health in laying hens
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Over the next decades, the world's population is forecast to increase by 25%, and to meet this challenge, food production needs to increase accordingly. Chicken Eggs constitute the most affordable source of animal protein available, resulting in consistent pressure on the poultry industry to optimize productivity and extend the laying period. Our approach takes advantage of the biology of laying hens, which, like humans, are diurnal and, therefore have a natural cycle of being active (and feeding) during daytime and sleeping during nighttime. Commercial hens are grown and maintained under standard ad libitum feeding conditions, which allows free and easy access to feed at all times. This management practice results in nighttime eating, which reduces the quality of sleep and disturbs their natural circadian rhythms in metabolic and other physiological functions. To follow the effect of nighttime eating exclusively, we have built an autonomic setup that can regulate feed availability and monitor nighttime eating events under standard farming conditions. Animals were randomly assigned to two cohorts of 20 hens, a control cohort under ad libitum conditions and a cohort whose feed availability is synchronized with the natural wake-sleep rhythm. Control animals were then monitored for eating events by the corresponding photoelectric sensors that are positioned parallel to the mangers. Our data show that synchronizing feed availability with the natural sleep-wake rhythm, improves egg production up to fivefold, from 15% in old controls to 80% in synchronized hens ($p = 0.0001$, t-test), improves the quality of sleep threefold ($p < 0.05$, t-test), and extends healthspan. Further, our setup uncovers that even though hens accumulate feed in their crop before nighttime, the average hen exhibits nighttime eating events every hour of the night. Taken together, we offer a simple environmental intervention to substantially extend the profitable lifespan of commercial flocks.

Session 86

Theatre 10

Effect of recycled litter on broiler growth performances

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In poultry farming, broiler poultry are raised in facilities with clean litter, normally composed of wood chips and sawdust, which involves millions of tons of resources annually. After the production cycle, these litters, dirty with bird excreta, constitute a biological risk and are discarded. Reusing litter material reduces environmental impact and production costs, but requires prior treatment to mitigate possible harmful effects on birds and compromising the productivity and safety of chicken meat. To reduce the microbiological risks of recycled beds, treatments are applied to inactivate residual microorganisms that undergo, for example, composting or the application of chemical products. The objective of this work was to evaluate the use of recycled litter, treated by composting, on the growth performances of broiler chickens. For this purpose, 240 male chicks of the Ross 308 AP strain were controlled, between zero and 24 days of age. They were housed in 30 pens with 8 birds, and divided into 2 treatments (15 replicates per treatment): control with clean litter (CL) or recycled litter (RL). Animals live weight and feed intake were evaluated at 0, 12 and 24 days of age and the average daily feed intake, average daily gain and feed conversion ratio were determined. A GLM analysis was performed with treatment as a variable factor and considered the Periods: 1 (P1; 0-12 days), 2 (P2; 12-24 days) and total (PT: 0-24 days). With the exception of the initial live weight and the conversion ratio in periods 2 and total, the other parameters worsened ($P < 0.05$) in the reused litter. At 24 days, the chicks in the control group had 100g more of live weight (1460 vs. 1360 g) and weight gain and feed intake increased by 4.3 and 7.2 g/day, respectively. In this work, recycled litter had a negative effect on poultry performances, and microbial inactivation methodologies should be improved or clean litter should continue to be used in broiler production. Acknowledgments: This work was supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Enhancing Chick Health: Exploring Egg Quality Variation and Maternal Performance in the Fayoumi Breed Raised in Tunisia

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Optimizing chick health and development from the egg stage is crucial to improving overall profitability. In this context, the objective of this work was to examine the relationship between the quality of the mother, that of her eggs and the performance of the chicks in the Fayoumi chicken breed raised in Gabès. To do this, 34 hens and 6 roosters were raised in a farmyard in spring 2021. Morphometric measurements were carried out on the hens, eggs and chicks. Additionally, blood samples were collected from the hens and chicks for hematological and biochemical analyses. Our results indicate that hen body condition was positively correlated with egg weight, length, width and volume, but negatively correlated with egg shape index. In addition, our results show that the body condition of the hen was positively correlated with the yolk weight of its eggs. These results suggest that higher quality hens produced heavier eggs with a higher proportion of yolk. In addition, our results reveal that the body condition of the chick at the age of 15 days was strongly correlated with its condition at hatching, which itself correlated with the size of the egg. In conclusion, our results show that good quality hens lay larger eggs which in turn produce better quality chicks.

Session 86

Poster 12

Lysozyme activity and viscosity of albumen and amniotic fluid of incubated eggs stored for up to a week

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The study aimed to analyze the enzymatic activity of lysozyme and the viscosity of albumen and amniotic fluid depending on egg storage. Hatching eggs (n=1200) of Rosa 1 hens were used, divided into 3 groups: fresh eggs (1) and eggs stored for 4 (2) or 7 days (3) and incubated for 21 days. Thin and thick albumen on day 0 and thick albumen and amniotic fluid on days 7 and 14 of incubation were collected (n=12 per group on the day). The viscosity (Brookfield DVNext viscometer) and lysozyme activity (spectrophotometric method and *Micrococcus lysodeikticus* bacteria) were analyzed. Hatchability was calculated. One-way ANOVA and Tukey's test (P<0.05) were used. The hatchability decreased with storage time. The lysozyme activity on day 0 in thin albumen was lower in group 3 than in the others (P<0.001), and in thick albumen, it was higher in group 1 (P<0.001). The highest viscosity of thin and thick albumen was found in group 3 (P<0.001; =0.010). On day 7, group 3 showed lower viscosity than group 1 (P=0.048), and on day 14, the viscosity of amniotic fluid from group 2 was higher than in group 1 (P=0.009). Higher lysozyme activity in albumen on day 7 was demonstrated in group 3 than in group 2 (P=0.005) and on day 14 – in group 3 than in the others (P<0.001). Higher lysozyme activity in amniotic fluid in group 2 compared to group 3 on day 7 was noticed, and on day 14 – the lowest activity was found in group 1 (P<0.001). The storage time influenced the albumen viscosity through the loss of water from the egg (evaporation); the lower lysozyme activity in the stored eggs could be related to the lower antibacterial protection, which increased significantly in 2nd week of incubation.

Changes in brain gene expression after in ovo stimulation by sodium butyrate during embryo development of chicken

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Butyrate, which belongs to the SCFAs, is produced by intestinal bacteria. Intestinal cells can use butyrate as an energy source. Butyrate has anti-inflammatory effects mediated by signaling pathways, such as modulation of pro-inflammatory cytokines through inhibition of NF- κ B activation. Recent years of research indicate that sodium butyrate improves poultry production and influences the immune response and microorganisms in the gastrointestinal tract, especially in the intestinal. Changes in the gut microbiota profile can affect gut-brain response in poultry. The brain and intestines participate in bidirectional communication with the help of the endocrine and nervous systems. The aim of the study was to analyze the effect of in ovo stimulation with sodium butyrate on gene expression level in the brain of broiler chickens. One thousand incubated eggs of Ross 308 broiler chickens on day 12 of incubation were injected with saline in the control group and with postbiotic sodium butyrate (SB; three doses: 0.1%, 0.3%, 0.5%) in the experimental groups. Brains were collected postmortem on day 42 after hatching from eight randomly selected individuals from each group for RNA extraction. The panel of genes included: immunity-related genes (IL1 β , IL2, IL4, IL6, IL8, IFN γ , IFN β), heat shock genes (HSPB1, HSPB5, HSPB8, HSPB9, HSP70) and other genes (BDNF, GSK3 β , GR, NR2A, NF- κ B p65). The results showed a decrease in expression for all genes, at different levels depending on the concentration of butyrate in the experimental groups. Gene expression was down-regulated in the dose with the highest concentration (0.5%). The 0.1% dose did not generate significant differences compared to the control. The most optimal effect was demonstrated by a dose of 0.3%, in which changes in the abundance of bacteria in the intestines contributed to changes in the gene expression level. The above results can be identified as an effect of the gut-brain axis in which SB directly affected the gut and indirectly affected the brain. It can be assumed that as the amount of research increases to understand the functioning of this relationship, it will be possible to reduce or eliminate poultry diseases in the future.

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Poster 14

Interaction of probiotic Bifidobacterium and Bacillus strains with intestinal cells in co-culture models for in ovo application

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In ovo technology enables controlled modulation of the host microbiome and immunity prior to hatch. The aim of this study was to optimize predictive modelling of these effects using probiotic candidates for in ovo treatment. The new chicken intestinal cell line Chic8E11 and the Caco-2 cell line (as a reference) were inoculated separately with probiotic Bifidobacterium and Bacillus strains or their culture supernatants, at a ratio of 10:2 (bacteria/supernatant: cells), and co-cultured for 24h at 37°C, with 5% CO₂ (3 replicates). Adhesion tests and qPCR gene expression analyses were performed. β -actin and GAPDH were the reference genes used for the latter. Adhesion of the probiotics to Chic8E11 was higher than to Caco-2. The Bacillus strain had a stronger adhesion ability to Chic8E11 than Bifidobacterium, but the Bifidobacterium strain adhered more strongly to the Caco-2 cells. For Chic8E11 cells, increased expression of villin, cytokeratin 18 and occludin genes was observed with the Bifidobacterium strain and the supernatants of both strains. However, direct contact with the Bacillus strain resulted in a decrease in the expression of these genes. For Caco-2 cells with Bifidobacterium, the expression of E-cadherin, cytokeratin 20, zonula occludens-1, occludin and claudin-1 increased. With the Bacillus, the expression of cytokeratin 18 and cytokeratin 20 increased, while E-cadherin, occludin, claudin-1 and villin decreased. Results obtained here will be used to translate in vitro findings to in ovo/ in vivo studies with the same probiotics, and further develop a predictive model for effects in the host organism. Financed by 2019/35/B/NZ9/03186 (OVOBIM) National Science Centre Poland.

synchronizing feed availability with the natural rhythm improves productivity and health in laying hens
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Chicken Eggs constitute the most affordable source of animal protein available, resulting in consistent pressure on the poultry industry to optimize productivity and extend the laying period. Our approach takes advantage of the biology of laying hens, which, like humans, are diurnal and, therefore have a natural cycle of being active (and feeding) during daytime and sleeping during nighttime. Commercial hens are grown and maintained under standard ad libitum feeding conditions, which allows free and easy access to feed at all times. This management practice results in nighttime eating, which leads to reduced quality of sleep and subsequently disturb their natural circadian rhythms. To follow the effect of nighttime eating exclusively, we have built an autonomic feeder that can regulate feed availability and monitor nighttime eating events under standard management protocol. Animals were randomly assigned to two groups of 20 hens, a control group under ad libitum conditions and a group whose feed availability is synchronized with the natural wake-sleep rhythm. Control birds were then monitored for eating events by the corresponding photoelectric sensors that are positioned parallel to the mangers. Our data show that synchronizing feed availability with the natural sleep-wake rhythm, improves egg production up to fivefold, from 15% in old controls to 80% in synchronized hens ($p = 0.0001$, t-test), improves the quality of sleep threefold ($p < 0.05$, t-test), and extends healthspan. Further, our setup uncovers that even though hens accumulate feed in their crop before nighttime, the average hen exhibits nighttime eating events every hour of the night. Taken together, we offer a simple environmental intervention to substantially extend the profitable lifespan of commercial flocks.

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Poster 16

Probiotic and prebiotic compounds as in ovo vaccine adjuvants

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The aim was to verify, whether probiotics, prebiotics, or synbiotics injected in ovo would improve the efficacy of an Infectious Bursal Disease (IBD) vaccine in broiler chickens. Two candidate probiotic and prebiotic compounds were selected in vitro by testing 12 prebiotics and 12 probiotics. In experiment 1, a Bacillus strain and vegetable protein hydrolysate were injected in ovo alone, and also in combination. In Experiment 2, Bifidobacterium lactis and Astragalus polysaccharides were injected in ovo, in a similar design. Probiotic was injected alone at a dose of 10^6 CFU and 10^3 CFU when applied with the prebiotic (1mg). In ovo injections (50 μ l/egg, 140 eggs/group), were performed to the amnion on day 18.5 of incubation. There were 4 groups in each rearing experiment: vaccine alone (V), V+probiotic, V+prebiotic, V+synbiotic (4 replicate pens, females only). After 4 weeks, the concentrations of IBD antibodies were measured using ELISA in the vet lab. Hatchability and growth were not affected. A significantly higher production of antibodies was detected in the V+synbiotic groups in both experiments as follows: Experiment 1: Bacillus with vegetable protein hydrolysate (1139B) vs. vaccine alone (126A); Experiment 2: B. lactis with Astragalus polysaccharides (67.5A) vs. vaccine alone (35.0A). In the V+synbiotic group, beneficial microbiota identified by qPCR (Bifidobacterium ssp. and Lactobacillus ssp.) were elevated compared to the other groups ($P \leq 0.05$). In conclusion, a single in ovo injection of a probiotic, prebiotic, or synbiotic has a potential adjuvant effect on vaccination and will be further explored as a novel prenatal immunization strategy. The study also confirms, that the potentially synergistic effects of the bioactive compounds can be optimized in vitro, before in ovo injections. NCN 2019/35/B/NZ9/03186.

Validation of a macroscopic classification at the processing plant of breast muscles affected by growth-related abnormalities through meat quality evaluation in broilers

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The present study was developed as preliminary research within the PRIN PACHOL4 project, which aims at investigating the molecular basis of growth-related abnormalities (e.g., wooden breast; WB) affecting the P. major muscle (PM) of fast-growing (FG) broilers and impairing the quality traits of affected meat. An accurate classification of PM is required to investigate factors ascribable to the onset of these abnormalities, thus avoiding misleading results. This work was thus performed to validate the macroscopic classification of PMs (as affected and unaffected by WB) to be further investigated. PMs belonging to three homogenous flocks (Ross 308 males 45-d old 3.2 kg of live weight; ADWG>85g) were collected at a commercial processing plant 3h post-mortem and classified relying on their macroscopic traits as normal (NORM, 30 PM/flock) and abnormal (ABN, severe WB; 30 PM/flock). Then, the main quality traits and technological properties were assessed: ultimate pH (pHu), color (L*, a*, and b*), drip loss, cooking loss, and shear force. Principal component analysis (PCA) and hierarchical cluster analysis (HCA) were applied using meat quality data to validate the sample classification implemented at the slaughterhouse. The effect of WB on the quality traits of chicken PMs (N=180; 30 PMs/group/flock) was tested by Student t-test using STATISTICA 10 (StatSoft). PCA and HCA analyses were performed in R environment (v.4.3.2). As expected, meat quality evaluation evidenced significant differences (P<.05) between NORM and ABN concerning the pHu, redness (a*), cooking loss and shear force. PCA allowed to explore the meat quality traits of NORM and ABN samples and identified pHu, lightness (L*), cooking loss, and shear force as potentially useful for sample classification. HCA showed sample clustering partially resembling the macroscopic classification implemented at the processing plant. Overall, the present results allowed us to perform a more accurate classification of PM samples to be selected for further analysis. The research was supported by MUR, PRIN 2022 PACHOL4 (Prot.n.2022EPWEPW).

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Poster 18

The optimal bedding for broiler health – characterization of various bedding materials for an optimal broiler environment

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To achieve optimum growth performance of growing broilers, reducing environmental contamination by microbes and pathogens plays an important role. One aspect that needs to be considered to minimize the risk of disease transmission is the provision of litter that prevents health and inhibit bacterial growth. Therefore, the aim of this study was to characterize different litter materials according to health-related criteria and determine the optimal range of trait values to reduce bacterial growth in order to develop practical indicators. A literature research was performed to identify all relevant bedding materials and health-related criteria. Various bedding materials (e.g., cereal straw, spelt husks, maize silage, wood shavings) were then bought to determine characteristics such as volume, grain size, dust content, pH value and buffer or water-binding capacity. In addition, nutrient fractions and degree of soiling were chemically analyzed. It is already known from the literature that bedding materials should generally have a high water binding capacity and buffering capacity. In addition, a low dust content is beneficial to prevent respiratory diseases, as well as a low pH value and degree of soiling reduces microbial growth and supports health. The results from the laboratory analysis will be presented. In conclusion, the study tested practical indicators were suitable to provide additional information to farmers and generate value for preventive health management. This work is financially supported by the Federal Ministry of Food and Agriculture, granted by the Federal Office for Agriculture and Food.

Periparturient supplementation of postbiotics on lactation performance in transition dairy cows

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This study aimed to evaluate the effect of postbiotics on lactation performance in dairy cows supplemented during the transition period until 60 DIM. Sixty Holstein dairy cows were enrolled in a randomized complete block design from -35 to 60 DIM and blocked according to expected calving day, parity, and previous lactation energy-corrected milk. At -35 DIM, cows were assigned to either a basal diet plus 100 g/d of ground corn and 25 g/d of placebo (CTL; n=30) or a basal diet plus 100 g/d of ground corn and 25 g/d of postbiotic (PB; n=30). Cows were fed 1X/d with TMR, and treatments were top-dressed after feeding. From -35 to -22 DIM, cows were fed a far-off diet; from -21d until calving, cows received a close-up. From calving to 30 DIM, cows received a fresh cow diet, and from 31 to 60 DIM, cows were fed a post-fresh diet. Data were analyzed using the MIXED procedure of SAS. Pretrial BW was greater (P<0.01) in CTL cows compared with PB; therefore, pretrial BW was used as a covariate. A trend (P=0.10) was observed for greater milk yield (47.4 vs 50 kg/d) in PB cows compared with CTL. Similar to milk yield, a trend (P=0.10) for greater protein yield (1.43 vs 1.50 kg/d) was observed in PB cows compared to CTL. The results suggest that supplementation of postbiotics can help to increase milk production; however, more research is needed to clarify the effects of this prototype on transition dairy cows' performance.

Session 87

Theatre 2

The influence of societal issues on representations of the breeder's profession and perceptions of change

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Today's major societal issues (environment, animal welfare, etc.) present livestock sector actors with a challenge: the need to adapt livestock farming practices to improve their sustainability. The multi-stakeholder Entr'ACTES project (CASDAR 2023-2026), led by the French Livestock Institute, proposes to analyze the impact of societal controversies on the practices of livestock industry players and their interactions with the rest of society. Four qualitative surveys were carried out in 2023 to assess the place of societal issues among the concerns of livestock farmers (cattle, pigs and poultry), the image they have of their profession, their perception of change and their need of support. Almost 60 semi-structured interviews were conducted in 2023 with livestock farmers and support staff (technicians, advisors) in France. Then, a quantitative survey of around 500 farmers completed and refined these results (survey in progress). Initial results show a diversity of representations of the breeding profession, societal issues and change by the stakeholders interviewed. The thematic analysis shows the climate of uncertainty and pessimism prevailing in the livestock sector, as well as the opportunities and challenges faced by farmers in this context. A typological analysis enables us to characterize the diversity of ways in which farmers think and act. The four breeder profiles thus identified show the extent to which societal issues are difficult to accept and share by some breeders, or conversely are widely integrated and taken into account by others. In addition, analysis of perceptions of change shows that, in order to drive the agroecological transition on farms, the use of change management tools in advisory services needs to be adapted to the different breeder profiles.

The need for tailored breeding goals by farm type and agro-ecological zone in crossbred dairy cattle in Oromia and Sidama, Ethiopia

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Dairy cattle in Ethiopia have been under the influence of crossbreeding practices since the 1950s, yielding variable results due to differences in genetic composition, management systems, agro-ecological zones, feeding practices and meteorological settings. So far, the breeding goal for dairy cattle in Ethiopia did not consider these components. We focused on disentangling the influence of environmental and management parameters from the genetic components by looking at interactions between breed and agro-ecological zone and farm type. Performance data of dairy cattle were collected on a weekly basis from 320 dairy farms in four zones of Oromia region and three zones in Sidama region between June 2022 and April 2023. Genotypes (100K) were collected for 848 animals (crossbreds of Arsi-Bale, Borana, Jersey and Holstein) on 295 farms and were aligned with reference populations of several European, Asian and African genotypes. Meteorological data was extracted using interpolation of nearby weather stations based on the farm's coordinates. Mixed linear models were used to find out how performance, agro-ecological zone, meteorological parameters and breed composition were associated. The optimal type of crossbred varied across agro-ecological zones and farm type in Ethiopia, meaning that breeding goals should be tailored to production system and agro-ecological zone.

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Theatre 4

Cow characteristics associated with lactation performance of cows with customized lactation lengths on commercial dairy farms

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Around calving, dairy cows have an increased risk for health issues. Some dairy farmers extend the voluntary waiting period (VWP) to extend the calving interval (CI) and herewith reduce the frequency of calvings. Moreover, farmers customize the length of the VWP for individual cows based on cow characteristics before insemination. In an earlier experimental study, models were developed where parity, peak yield, and milk yield (MY) and fat content in the week before insemination were associated with MY end lactation and MY per day of CI (MY/CI). The current study aimed to evaluate these models on dairy farms. Over 3,000 complete lactations of Holstein Friesian cows between 2014 and 2019 from 13 farms were grouped by parity (1 or 2+) and calving to first service interval (CFSI; < 84 d, 84-140 d, 140-196 d, 196-252 d, and ≥ 252 d). Lactation curves were fitted with a mixed model by using a Wilmlink curve with a negative gestation effect to derive MY end lactation and MY/CI. Parity, CFSI, their interaction and MY before successful insemination affected MY end lactation ($R^2=0.54$). A similar model was created for MY/CI with addition of peak yield and fat content before successful insemination ($R^2=0.87$). When including farm as fixed effect in the final models, these R^2 were 0.79 and 0.89 respectively, indicating an additional effect of farm on MY end lactation. Overall, a greater MY before insemination was associated with a greater MY end lactation, and a greater peak yield and MY before insemination and a lower fat content before insemination were associated with a greater MY/CI, indicating that cows with these characteristics may be suitable for an extended VWP.

Impact of Inbreeding on Dairy Cow Survival

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The dairy industry strives for sustainable practices, focusing on dairy cow health and longevity. However, inbreeding poses a significant challenge to this as it affects various traits negatively. This study investigates how pedigree (FPED) and genomic (FROH) inbreeding impact the survival of dairy cows post-first calving. Cox proportional hazards regression analysis on 56,309 Danish Jersey (JER) and 103,716 Danish Holstein (HOL) cows was used to estimate Hazard Ratios (HR) on different covariates. In the analysis, time was the cow's lifespan from first calving until culling or to the data extraction date (censored records). FPED or FROH were used as continuous covariates in the analysis. Additionally, the analysis was performed across different runs of homozygosity (ROH) length categories: 1-2 Mb, 2-3 Mb, 3-5 Mb, 5-10 Mb, 10-17 Mb, and >17 Mb. Results indicated that overall, JER cows exhibited a lower HR for culling compared to HOL cows. We observed HR increases of 1% per FPED and 2% per FROH. Statistically significant HR coefficient increases were seen within ROH length categories 2-3 Mb, 5-10 Mb, 10-17 Mb, and >17 Mb. The largest HR coefficient was in the 2-3 Mb ROH category, with a 4% increase per Froh unit and the highest HR for mean Froh was in the 5-10 Mb ROH category, at a mean HR of 4.3%. No significant differences in HR were found between breeds within different ROH categories. In conclusion, inbreeding slightly increased HR, and higher HR was seen for the longer ROHs, which indicates that recent inbreeding is more harmful to dairy cows.

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Theatre 6

Investigation of culling of primiparous cows in Swedish herds

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Swedish dairy cows have an average lifespan of 5.1 years. To increase longevity should be aspired, both for environmental and economic reasons, as that would increase the proportion of time the cow spends as a productive dairy cow compared to the recruitment period as a heifer. However, to increase productive life, further insight on reasons for culling during first lactation is needed. The objectives of this study was to describe the most common reasons for culling in primiparous cows for the two most common breeds in Sweden, the Red Dairy Cattle (RDC) and Holstein (HOL) cows. Data from the Swedish National Herd Recording Scheme on calvings, cullings, inseminations, and test milkings were collected from 4023 primiparous RDC and HOL cows, having their first calf in 20 high-yielding commercial dairy herds in the south of Sweden. Culling reasons were categorized into 12 categories. Comparisons of culling incidence, reported as the proportion culled of all cows included, between breeds were performed with Chi-square analysis and for each culling reason category with Bonferroni correction to account for multiple comparisons. Our preliminary results show that a total of 22% out of the (894/4023) included cows were culled during their first lactation. The overall culling incidence was higher for RDC than for HOL cows (20 vs 26%, $P < 0.0001$), and the most commonly reported culling reasons was Impaired fertility followed by Low yield. To conclude, both total culling incidence and culling reasons differed between the two main Swedish breeds.

The diversity in dairy cattle reticuloruminal temperature: Exploring the influence of temperature humidity index

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As a response to escalating global temperatures, there is an increasing reliance on climate factors to predict and assess the impact of heat stress on dairy cattle. The temperature humidity index (THI) accounts for the combined effects of environmental temperature and relative humidity to assess the risk of heat stress. However, as a herd level prediction, this approach negates the individual animal responses seen with changing environmental conditions. We assessed the variability in reticuloruminal temperature (smaXtec Animal Care GmbH) data for 1,429 cattle (mixed breeds) on three dairy farms across four years. We then compared it to interpolated THI data from surrounding weather stations and computed daily maxima (THImax). To avoid spurious associations between THI and reticuloruminal temperature data, analysis included both raw and residual data (obtained after removal of seasonal and long-term trends) in the development of linear mixed models. Our findings reveal a greater average reticuloruminal temperature deviation (°C) across individual farms at a THImax of 67, with a marked increase when THImax reaches 75. Assessment of reticuloruminal temperature deviation in response to THImax deviation reveals a unique response for each individual animal. Analysis of the slope, which represents the rate of increase in temperature with increasing THImax, could therefore provide the first step for the development of a core-body temperature-derived phenotype for the selection of heat-tolerant animals.

Compost-bedded pack systems: the importance of tilling on manure microbial populations

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Compost-bedded pack (CBP) is a common housing systems in northeast Spain. This system performs a tilling process upon manure in order to use it as bedding material for the cattle, which may alter the structure of the manure microbial ecosystem. This impact could differ between depths and be differently affected by temperature changes along the year. Manure samples (n=55) were taken at 0 cm (Sup) and 30 cm (Deep) depth from the composted bed during two seasons, winter (Win) and summer (Sum). Samples were immediately frozen and genomic DNA was extracted. High-throughput sequencing of the V3-V4 hypervariable region of 16S rRNA was conducted on Illumina MiSeq system. Taxonomic assignment of phylotypes was performed using a Bayesian Classifier trained with Silva database; both microbial alpha and beta diversity as well as the taxonomic profiles of the samples were analyzed using “vegan” and “mixomics” packages from R software (v3.6.3.). Alpha biodiversity did not present significant variations among depths or seasons in terms of Shannon and Simpson index (P>0.05). Sup layer had a higher number of species; nevertheless, the number of dominant species was lower at that layer (P<0.01). Community structure was clustered by both season and depth (P<0.01), although separation between seasons was more remarkable in Sup given that temperature in Deep layer remains more stable along the year. Deep layer doubled population of Euryarchaeota phylum (P<0.01), related to methane-forming archaea, which also increased significantly in Sum samples. Results show the differences in microbial structure and composition on the layers generated in composted manure of CBP. Moreover, they highlight the importance of environmental temperature on microbial activity associated with potential GHG emissions in manure.

Use of a Quality Cost Delivered approach in dairy cattle farm

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In dairy cattle farms most part of the effort is devoted to technical choices, with lack of view in the managerial approach and the systemic vision of the company. Quality Cost Delivered (QCD) is a managerial approach that had origin in automotive sector. This approach addresses the extent of quality, cost and time needed for a given process. In dairy cattle farms feed costs is the most relevant and need to be analyzed in a holistic approach. The aim of this study (set within PNRR Agritech 5.2.6;CUP J83C21000300006) was to adapt QCD approach to analyze the feeding area of 2023 in a dairy cattle farm of 217 milking cows in Arborea (Italy). For quality measurement a parallel conceptualization was made: farm operators were assumed as service providers and cows as consumer. For operators, quality was measured in “defeat of the service”, and its Key Performance Indicators (KPI) was the numbers of time that a problem interrupted or retarded the service (e.g. fault or no planned end of stocks). For herd, quality was measured in “customer satisfaction” and its KPI were farm Feed Efficiency (FE), Income Over Feed Cost (IOFC) and % of times that milk quality was not conforming to high quality grid of payments (HQ). Costs included feed cost of purchased and self-produced feeds at market value, labor, fuel, maintenance cost were expressed in €/cow in milk. Time was collected by work sampling (WS) and then expressed in time/cow in milk and classified as: waste, with value and with non-added value. For the operators’ quality, defeats resulted once in a 13 days window. Herd FE was 1.02, IOFC 444,187 €/yr and 16% of milk quality analysis were not conformed to HQ over the year. Total feeding costs were 1,147,148 €, being 95% feeds, 2% operators, 2% fuel and 1% maintenance. When expressed per milking cow, feed and other costs were 13.72 and 0.72 €/d, respectively. About labor time, 1 operator was needed, working daily for 7 hours (21 min/d and 121 hours/yr per cow in milk). Time breakdown from WS was 31% waste, 7% non-added value and 52% added value. In conclusion, this approach could be applied in strategical planning of dairy farms to monitor routine process.

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Theatre 10

Milk haptoglobin detection using chemiluminescence imaging with smartphone camera

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Bovine mastitis (BM) is one of the most frequent diseases in dairy cattle, having a large adverse effect on farm economics, including increased treatment costs, decreased milk yield, escalation of somatic cell counts, increased risk of removal from the herd or even death. Early detection of new BM cases is of high importance for infection control within the herd. Traditionally, BM detection depends on the efficacy and reliability of conventional techniques, which are designed to measure somatic cell counts (SCC), detect causative pathogenic bacteria, and reveal inflamed status associated with secreted biomarkers. The latter are secreted into plasma or milk upon inflammation, infection or trauma and are used as predicting analytical biomarkers for clinical status assessment. Herein, we utilize enhanced chemiluminescence (ECL) imaging by digital imaging (smartphone) for rapid and sensitive detection of whole milk samples quality and safety, outside laboratory boundaries. The system allows routine bio-diagnostics of haptoglobin (Hp), a predictive acute phase protein of BM, by utilizing the binding capacity of hemoglobin (Hb). The CL intensity is linearly proportional to Hb-Hp complex formation, resulting in peroxidase-like activity inhibition of the luminol-H₂O₂-Hb ECL system. Moreover, the influence of different SCC and pathogen types on the secreted milk Hp levels obtained from Holstein cows was established, in agreement with a commercial assay kit. The proposed ECL sensing concept offers a cost-effective, portable, simple, label-free and reliable systematic analysis of Hp biomarker for BM, potentially initiating a positive effect on animals’ health and the overall economy of dairy farms.

Infrared thermography in dairy goats to define health traits – preliminary results

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Infrared thermography (IT) is a non-invasive technique, which can be used as disease indicator or as measurement to detect heat stress (Tattersall, 2016). In dairy goats this technique might serve as a method to define health traits on-farm and with this serves as an objective evaluation tool. The present study presented preliminary results of a total of 137 dairy goats examined by IT in three perspectives each: left side, right side and back view. IT images were used to generate six temperature variables: eye, body, front leg, hind leg, crown hem and udder. Additionally, faecal egg count (FEC), age, weight and milk yield were tested as fixed effects (using SAS Studio). Age resulted in a negative correlation to IT body and IT udder temperature, with younger animals having higher IT-temperatures. Interestingly, FEC is correlated to IT leg temperature, with animals having a higher FEC resulting in lower IT-temperatures. Further examinations and image evaluations are necessary to generate more data in order to define objective health traits. The outdoor climate is included in the following examinations to evaluate effects on IT images in more details. Additionally, IT horn temperature will be analysed in more detail to gain information about thermoregulatory purposes. Tattersall, G. J. Infrared thermography: A non-invasive window into thermal physiology. *Comp. Biochem. Physiol. A. Mol. Integr. Physiol.* 2016;202:78-98 The present research is funded by the German Federal Ministry of Food and Agriculture (funding code 28N-2-035-02).

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A novel hydrogel system for ambient transport of bovine germplasm

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Bovine germplasm collection is well established and widens access to genetics to support sustainable cattle production as the world aims for Net Zero. Conventional transport of bovine germplasm, under liquid nitrogen, can be challenging; especially in areas lacking a robust cold-chain network. Here, TissueReady™ hydrogel systems (Aterlix, UK), initially designed for transporting human stem cells, were tested for bovine germplasm transport. Preliminary data revealed incompatibility between bovine semen extenders and the hydrogel system, giving poor spermatozoa (spz) survival. Here, changes were made to reagents to support spz survival. Semen samples were collected and divided between controls and modified Hydrogel Kits (HG) then stored at ~17°C. Motility (mot) and progressive motility (prog) were assessed at set storage times. At T0 the control was 61.5% mot and 46.3% prog. At T5 HG samples had higher motility results than controls (HG – 44.23% mot and 27.23% prog; control – 22.6% mot and 12.2% prog). T72 motility parameters decreased for all samples although HG samples were higher (HG 12.4% mot and 1.78% prog; control 2.8% mot and 0.6% prog). A second study investigated embryo storage in standard TissueReady™ kits. Once released from kits, after up to 3 days storage at 4°C, embryos continued to develop with standard culturing. In summary, modified TissueReady™ hydrogel systems show the potential to extend the fresh shelf life of bovine germplasm to several days, enabling transport without the use of liquid nitrogen. This is particularly useful for remote areas/sites with limited cold-chain infrastructure. Further work is required to establish the limits of germplasm storage.

Water Quality: Manganese in drinking water for dairy cattle

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Climate change affects livestock systems through direct impacts on the physiology, behavior, production and welfare of animals and indirectly through the availability, composition and quality of food. Water is essential for the life of animals, since it intervenes in various metabolic processes. An inadequate water supply could reduce the health and performance of the animals. In dairy farms, the use of quality water is essential to maximize the milk production of animals. The aim of this study is to see if water quality (excess manganese) affects production, reproduction and health in dairy cows. The study was carried out on a dairy farm in Alentejo-Portugal. The objective was to verify whether there were any effects of manganese toxicity. Two experimental groups were established, balanced for: Age, Number of calvings, Milk butyric content, Milk protein content. All the experimental groups were in the same conditions: Farm/stable, feed, ambient temperature and humidity, same number of drinking troughs, same trough space, same feed distribution time, same farm health management. Variation factor – manganese concentration: Treated group <50 µg/L (DGAV, 2014), Untreated group >1500 µg/L. Blood, urine and milk samples were taken over a 4-month period. The preliminary results show that the group of animals without treated water (excess manganese) has a 22% reduction in the consumption of kg of feed and 26% in water consumption; – 879 litres/lactation; -2.8L/cow/day; + 63 days of drought; + 2.4 inseminations; + 57 days of calving interval; + 33% embryonic mortality rate; +19% use of oestrus synchronization protocols.

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Systematic review of serious games on livestock and how they consider sustainability

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Serious games seem to be used more and more in agriculture and especially in livestock. They are supposed to be useful for learning purposes but also for co-design or to evaluate decisions with simulation models. Thus, they should be used in sustainability issues. We realized a systematic review of published articles on the use of serious games to address sustainability issues of livestock. A PRISMA method have been used to make the review. We produced a corpus description of articles, a descriptive analysis of games and an analysis of assessment done on games. We borrowed also a framework developed to define what is sustainable agriculture in order to study how serious games can be considered (or not) as a tool for sustainability in agriculture. The results (based on 107 empirical studies) showed that serious games on livestock have been gaining in importance since 2019. They are seen as an effective means of enabling stakeholders to work together and tackle complex issues in a systemic way to achieve sustainability in livestock sector. Indeed, sustainability issues are increasingly taken together in games with more goals, strategies and field of action. Games are designed for learning but also to co-design new systems. This review provides a solid basis for future studies of serious games in livestock. Nevertheless, the weakness of the assessment carried out in the articles and the diversity of games does not make it possible to highlight the real impact of games which lead us to ask for a research agenda on this theme.

Heat stress effects on milk yield, milk components and udder health in early lactation dairy cows

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The aim of the current pilot research was to evaluate the effects that heat stress has on milk yield, milk components and somatic cells count (SCC), during the first 100 days in milk (DIM). A study-herd 40 Romanian Black and White dairy cows was set-up, 20 cows calved in early-summer (June-July), and 20 calved in early-spring (March-April, controls). Cows were evaluated for milk production using the ICAR 28AM+PM recordings, and received identical daily feed rations, with individual milk samples taken to determine milk components (fat, protein and lactose) and SCC. Milk production was corrected for parity, while data was computed as mean \pm SEM, using the non-parametric Mann-Whitney U test to estimate differences between calving groups. Milk yields and milk fat content during 100 DIM were significantly higher ($P \leq 0.05$) in cows that calved during spring, compared with cows that calved during summer season, under heat stress conditions. Calving season had no effect ($P > 0.05$) on milk protein or lactose content. Contrary to our expectations, the SCC levels were significantly lower ($P \leq 0.05$) in cows that calved in summer, these results being potentially attributed to environmental factors, such as lower humidity levels, and thus a lower pathogen load. The preliminary results suggest that calving season, aggregated with heat stress exposure during the transition period, have negative effects on milk yields and on milk fat content. However, the SCC expressed lower levels during the summer months early lactations, potentially due to environmental factors such as humidity and bedding hygiene.

Seasonal Patterns in Dairy Cattle Pregnancy Rates under European Temperate Conditions

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Heat stress has been shown to have a negative influence on fertility in cattle, leading to a negative impact on the overall reproduction efficiency in commercial dairy farms. However, limited research has been carried out to-date on the subject under temperate climatic conditions, in regions with all four seasons exhibited. Therefore, the aim of the current study was to evaluate the season's effects on pregnancy rates in dairy cows reared under temperate conditions. The study was carried out at the Research and Development Institute for Bovine Romania, where a herd of 110-118 purebred Romanian Black and White (Holstein strain) dairy cows and replacement heifers were managed and fed under identical conditions year-round. Data of 465 artificial inseminations (AI) was followed, and the resulting number of confirmed gestations for both multiparous and nulliparous cows, between 2022 and 2023, with a balanced number of AIs per season. For adult cows, pregnancy rates varied between $35.56 \pm 5.07\%$ in summer and $56.96 \pm 5.61\%$ in autumn, with significant differences ($P \leq 0.01$) being observed for summer, when compared with autumn and winter. Nulliparous cows were negatively impacted to a lesser extent ($P > 0.05$), potentially due to lower metabolic pressure and different diets, with younger animals being thus able to better thermoregulate, unlike adult lactating cows, which received feed rations significantly higher in energy, producing additional heat during the digestive processes. Regardless of the age group, a tendency towards lower pregnancy rates during summer season was observed, thus farmers and veterinarians should avoid practicing AI during intense heatwaves even in temperate climatic regions.

Goat grazing restores plant diversity in an abandoned field by inducing photo environmental heterogeneity
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Field abandonment induces encroachment by weedy plants and subsequent overgrowth, reducing biodiversity and ecosystem services. Appropriate management measures should be implemented to restore such abandoned fields. We evaluated the impacts of goat grazing in abandoned fields on plant diversity via changes in the photo environment over five years. Three conditions with three replicates each were implemented: grazing with three goats (GG), mowing with a bush cutter (MB), and no weeding (CON). Before grazing began in the first year, the entire experimental area was clear-cut using a bush cutter. Grazing was implemented for approximately 7–10 days each during spring, summer, and autumn annually. Weeding with a bush cutter was performed on the same day at the start of each grazing session. We set 6 permanent quadrats in each replicate, then identified plant species, measured the height and coverage of each species, and estimated the Shannon index (H'). The relative photon flux density (RPF_D) was measured before, immediately after, and one month after grazing and weeding in each quadrat. A generalized mixed model was applied to plant height, number of species, and Shannon H' index. Average plant height was highest in the CON field and lowest in the MB field. The interquartile plant height range in GG fields increased with the years. RPF_D was consistently lower in CON field than in GG and MB fields, whereas the difference in RPF_D between GG and MB fields increased with time after each treatment. The number of plant species was higher in GG than in MB and CON fields. Shannon H' was higher with GG and MB fields than with CON field. Goat grazing increases the number of plant species and improves botanical diversity by creating heterogeneity in vegetation height and photo environment.

Changes in milk constituents in dairy cows fed diets with high concentrate content

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High quantities of starch in diets for dairy cows may result in a decreased rumination, salivation, and in an increased short chain fatty acid production, causing a repeated moderate drop in rumen pH on a prolonged period of time. This ruminal pH depression is termed subacute ruminal acidosis (SARA). Signs of SARA include a decrease in dry matter intake, laminitis, milk fat depression, loose faeces, abscesses in the liver. SARA is difficult to diagnose. With the aim to identify suitable markers to diagnose SARA, we conducted a study with 23 dairy cows in mid-lactation kept in a free-stall barn, with an automatic milking system. The study lasted 63 days, comprising 3 periods (P) of 21 days each. During P1, cows were fed a control diet (52:48 forage to concentrate ratio; CONT1), followed for P2 where cows were fed a SARA inducing diet (35:65 forage to concentrate ratio; SAR2), to be then fed (P3; CONT3) the control diet. Samples collected included milk, rumen fluid, blood, urine, faeces and saliva. In this report, we present data of some milk constituents. No differences were detected in milk yield between CONT1 and SAR2. The content of fat, and the fat:protein ratio, were lower in SAR2 than in CONT treatments. The proportions (of the total fatty acids; FA) of saturated (SFA) and that of unsaturated FA (UFA) were similar among treatments. The ratio SFA:UFA was lower, and the proportion of C18:0 was higher in SAR2 than in the CONT treatments. Unexpectedly, the content of urea-N was highest in SAR2 (17.5, 19.6 and 17.0 mg/dl for CONT1, SAR2, and CONT3, respectively).

Rumen fermentation parameters prediction model using mid-infrared spectra of rumen fluid collected from Simmental dairy cows

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Rumen volatile fatty acids (VFA) are of paramount importance for energy supply to animal and their evaluation provide information regarding fermentation efficiency and rumen health. Mid-infrared (MIR) spectroscopy assay is rapid, nondestructive, inexpensive, not require reagents, and the results are reproducible. Thus, the aim of this study was to develop MIR calibration models for estimating the content of VFA (acetic, propionic, butyric, valeric, isovaleric, hexanoate and isobutyric acids), lactate, and urea in rumen fluid from mid-lactating Simmental cows. Rumen fluid samples were collected from 34 cows at 3 time points every two weeks during summer. Samples were centrifuged and the supernatant was harvested for MIR (Milkoscan FT2), VFA by GC, and ammonia and lactate by ILAB650 analysis. The spectra were subjected to Partial Least Squares regression analysis. Absorbances in the 4,911.2 to 4,303.2 and in the 4,189.2 to 2,897.2 cm⁻¹ range were considered. The optimization of the number of latent variables in the PLS model was made by leave-one-out cross-validation. The model quality assessment was based on the Root Mean Square Error (RMSE) and the correlation coefficient (R₂). Results suggested that the predictive model well-fits the data for some parameters. The R₂ (and RMSE) values for prediction of VFA as mmol/L and mmol/100mmol of total VFA were 0.82 (3.88) and 0.97 (0.02) for acetate, 0.97 (1.6) and 0.97 (0.02) for propionate, 0.95 (0.99) and 0.96 (0.001) for butyrate, 0.85 (0.1) and 0.86 (0.001) for isobutyrate, and 0.72 (0.11) and 0.71 (0.001) for hexanoate, respectively. The models also provided good predictions of pH (R₂=0.94, RMSE=0.1) and ammonia (R₂=0.83, RMSE=2.65). MIR spectroscopy models provided good predictions of rumen pH, VFA, and ammonia of rumen fluid, providing a reliable, simple, and rapid method to estimate rumen health and metabolism efficiency.

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Theatre 1

Livestock farmers knowledge and attitudes regarding agroforestry planning and management

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Silvopasture systems (SS) have been highlighted as a sustainable tool to achieve Net-Zero targets set out for the UK. In this project we aimed to explore the knowledge and attitudes of UK livestock farmers regarding SS planning and management. Farmers (n=48) answered an online questionnaire about the topic. The majority of participants were male (83%) and over 50 years old (50%). 55% said they didn't have the information necessary to plan and manage SS. They had similar intermediate knowledge (3/5) on the specific aspects of information to plan a SS implementation, but different knowledge levels towards SS related management practices (2/5 for pruning – 4/5 for grazing management). Most participants considered it important to receive technical support (71%) however, 74% had low expectations of getting it from governmental agencies, and 62% were eager to improve their knowledge and skills towards SS planning and management practices. 83% of participants are already getting information about these topics mainly through internet and research centers sources (31% for both). Profitability (56%), benefits to the environment (42%) and animals (39%) were cited as key for a successful SS. In conclusion, participants don't have sufficient expertise to plan and manage SS successfully; but they are willing to improve their knowledge and skills.

Environment as a challenge for intensive livestock farming

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The EU has high ambitions: improve quality of water (related to P and N leaching; max 50 mg nitrate in upper ground water) and Nature 2000 areas (related to ammonia deposition); overall 90% GHG emission reduction in 2040 compared to 1990 (Green Deal); more biodiversity (4% of each farm area set aside); pesticide use to be reduced by 50% in 2030. Especially in intensive livestock regions, N-leaching has been a dominant environmental aspect to deal with. In the Netherlands, it resulted in 1990 in founding of experimental farm De Marke, fully devoted to this topic. Countrywide, use of N has since then reduced from on average 400 kg to a max of 200-250 kg/ha these days. More recently, ammonia and GHG emissions are a hot topic. The choice is between extensification (is stimulated) and technical innovation. Extensification can be achieved by transforming to organic or regenerative forms of agriculture, of which examples will be presented, although the business model is under question. Regarding technical innovation, we search for integrated solutions: practices that combat GHG emissions more times increase ammonia emission, and vice versa. The potential of reducing ammonia emissions with innovative practices and techniques are being evaluated in case control studies. A list of floor types and techniques, like cow-toilet and Lely Sphere, are available with the established emission factors. In addition, we study the possibility of simultaneously filtering methane and ammonia from the air in housing facilities. The relatively low levels of methane concentrations in open barns complicate the removal by air filtering. Experiments with alternative ventilation techniques are ongoing to collect and concentrate methane from close to the mouth of animals. In general: Experience learns that results achieved in experiments are not automatically copied in the field, and that the effect of a technique or practice needs to withstand complaints by action groups submitted to the Juridical Courts. A national policy shift is envisaged from means regulations, based on prescribed practices and techniques, to purpose/target regulations, allowing the farmer to apply his own approach and capacity to reach the prescribed target.

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Theatre 3

Climate adaptive solutions? Public views on dairy cattle heat stress management strategies

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Heat stress, exacerbated by climate change, threatens animal welfare and questions how the dairy industry adapts to sustainability challenges. Gene editing (GE) for heat resilience, indoor shelter, and water sprinklers are strategies proposed to protect cattle from heat stress. However, these strategies may be associated with concerns which could threaten public acceptance; for example, GE is often associated with concerns around unknown consequences, indoor housing with concerns around lack of pasture access, and sprinklers with concerns around water resource use. Using a mixed methods factorial survey with 930 North American participants, we assessed the effects of these strategies on public acceptance of dairy farming scenarios and perceptions of the standard of care provided to cows and of the cows' quality of life in these scenarios. On a 7-point scale, scenarios using indoor shelter were less accepted (-0.31 ± 0.11) and perceived to provide a lower standard of care (-0.42 ± 0.11) and quality of life to the cows (-0.38 ± 0.11) than scenarios not using this strategy. Scenarios using water sprinklers were more accepted ($+0.35 \pm 0.11$) and perceived to provide a higher standard of care ($+0.48 \pm 0.11$) and quality of life to the cows ($+0.34 \pm 0.11$) than scenarios not using sprinklers. Use of GE, alone, did not influence these three outcomes. Qualitative responses supported the quantitative results and revealed a more nuanced understanding of how different strategies provided trade-offs in values and opportunities to balance these. Our results suggest that participants placed value on heat stress management strategies directly related to how animals are cared for by farmers over technical solutions like GE, and participants favored balancing trade-offs between multiple strategies.

Cell-based food: Promises and reality

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While there is consensus on the challenges facing agriculture, food and the environment, the innovations being developed to address them are diverse. Among these, cell-based food is a subject that poses significant technical, ethical, regulatory and commercial challenges to bringing these products to market. Although the culture of muscle cells is well known, many technical and economic obstacles still need to be overcome before large-scale production can be achieved. Although costs are falling, they are still too high to be competitive. Scientists in academic research are calling for precise information to be shared in the interests of transparency, especially on the characteristics of culture media and products. It is argued that these products cannot be called “meat” from a biological, semantic and legal point of view. When it comes to product characteristics and sustainability, few academic research studies are available or conclusive. The issue of animal welfare is also central, as is the potential consumer acceptance of cell-based foods, which is complex due to the interaction of many factors. To become a credible alternative, cell-based food must offer a real and proven added value over meat. This is not (yet) the case. One scenario would be the failure of its development due to unfulfilled promises by private companies, lack of support from public authorities and from consumers. Another scenario is the opposite, based on more investments, technological progress and support. It is likely that cultural factors (such as the culinary history of each country) will be among the key factors of choices between these scenarios.

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Challenges related to the introduction of the EU Green Deal in Poland

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In recent years, the dairy market in Poland has undergone significant changes. Market dynamics, growing consumer demands, and climate challenges influence the future of the milk sector. The background to all changes in Poland are changes in Europe and the world. Poland is one of the leading milk producers in the European Union with decreasing cattle population and growing milk yield. Central Statistical Office estimates that in December 2022, 174,706 herds of dairy cows were reared in Poland, i.e., over 6,000 less than in June. Among over 174,000 dairy farms only 18,066 control the milk yield. About 40% of milk production comes from the controlled farms. Introducing the EU Green Deal – a roadmap to sustainable economies poses many challenges to dairy breeders and producers. Although farmers agree on the need to take action to create sustainable economies and protect the environment, most are not ready to introduce changes in their farms at the expense of income. Farmers' protests are an expression of concern though the implementation of some Green Deal elements may be crucial for the development of the dairy chain. They expect systemic solutions, taking into account support from the state. Until today the EU programs and related financial resources improved the living conditions and economic condition of most residents but did not guarantee the basis for lasting and sustainable development. The Green Deal, properly implemented, i.e. respecting the traditions and customs of Polish farmers, may be an opportunity for the development of Polish agriculture. It is both a choice and a necessity. The direction given by the European Green Deal is good for farmers, consumers, and the environment. The Green Deal could be the answer to some rural problems, but it lacks good promotion and understanding. The presentation will discuss the state of dairy farming in Poland compared to the situation in Europe. Project CCCfarming National Centre for Research and Development (SU-SAN/II/CCCFARMING/03/2021)

Changes of Eurobarometer as a tool for collecting EU citizens' opinion on animal welfare

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Through Eurobarometer, the European Commission conducted four surveys (2005, 2007, 2016, 2023) to evaluate EU consumers' attitudes towards animal welfare (AW). To determine the main changes in question frameworks across the questionnaires and in the overall respondents' interest in AW, questions were grouped into those profiling consumers or collecting their opinion on AW and food of animal origin (AO). The first group was further divided into questions on either personal objective details (e.g., gender, age, education, occupation, family, residency, technological belongings) or personal opinions and behaviours (e.g., political views, religion, level of satisfaction with politics and economics). From 2005 to 2023, questions increased overall (25 to 33) and within the consumer profiling group (12 to 19). The personal objective details asked were rather consistent, both in number and content. Meanwhile, questions asking for personal opinions and behaviours increased greatly (1 to 10), mainly due to new inquiries about politics, economics, and personal satisfaction. Over the years, questions on AW and food of AO were consistent in number and general topics (i.e., opinion and knowledge on AW, food labelling and purchasing habits). However, they became more detailed and extensive, inquiring also on specific farming practices, transportation, and slaughtering, and on companion and fur animals. Furthermore, questions asking the same thing were formulated differently, making comparisons across years hard to draw. On average over the surveys, respondents represented the 0.007% of the over-15 EU population. Their opinion on AW remained steady both when considered important and not important, with a slight decrease of respondents regarding it as very important.

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Theatre 7

Testing feeding levers to decrease the carbon footprint of dairy farms

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Agriculture contributes to global greenhouse gases (GHG) emissions, particularly through methane and nitrous oxide emissions. In France, this amounts to 21% of the overall national GHG emissions (CITEPA, 2021), with around 10% attributable to ruminants. Thereby, dairy production is facing a challenge to reconcile food challenge, competitiveness, and environment. Many practices are identified in bovine production to reduce the GHG emissions and improve profitability. The dairy experimental farm of Trevezet (Brittany) implemented in 2019 a holistic experiment to build a low carbon footprint system and assess its technical, economic and environmental impacts (Brocard et al, EAAP 2022). Among the solutions tested to improve the system, 4 experiments were led to assess feeding techniques or ingredients: the addition of a commercial additive into the winter diet, the replacement of part of the maize silage by fodder beets, the implementation of a maxi-autonomy diet based of grass silage and ear-corn silage and the replacement of part of the protein concentrate by grazing of legume-rich swards in summer. These diets show limited impacts on the GHG emissions or the global Carbon footprint of the farm but may have negative impacts either on the dairy production (the maxi autonomy diet, summer grazing in case of drought), the margin over feeding cost (cost of the additive, of the maxi autonomy diet), environmental impacts (high frequency of pesticides for fodder beets) or on the workload. Some may have positive features like a diversification of the forages to face global warming (fodder beets). These 4 levers related to dairy cows' diets will be benchmarked from technical, economic and environmental point of views.

The effect of dairy farming system on GHG emission estimations in Latvia pilot farms

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The structure of dairy farms in Latvia is influenced by historical changes at the end of 1990. There was time to make decision on the management system of land. Before collectivization in 1940 average farm size was 30 ha, in Latvia. Nowadays, the structure of dairy farms has changed, but still stays heterogeneous, in Latvia. This research aims to choose and monitor pilot farms that representing different dairy farming systems to collect reference data based on actual farm characteristics for the estimation of emission for greenhouse gas (GHG) inventory. Data monitoring plans were created based on IPCC general guidance for collecting data and adapting data for inventory use. First, to identify the management practices farms were categorised by management system as conventional or organic farms. Secondly, the productivity level of cattle by different breeds was considered. Thirdly, the weight of animals of different ages was determined as important outcome of monitoring as GHG calculations must include proper animals weights. Data collection is a key part of the national inventory arrangements, therefore based on collected data it will be possible to estimate emissions levels for different farming systems depending on local circumstances. Furthermore, it is important to extend further research according to the farm system, to provide a holistic picture of the emission profile of different farming systems, and implement appropriate mitigation strategies for dairy sector that is the biggest source of emissions in Latvian livestock sector.

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Assessing the climate impact of Norwegian dairy production

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The research based HolosNor family of whole-farm models are designed for estimating calculations and mitigation options of CO₂-eq at Norwegian farms. HolosNorDairy are developed for combined milk and meat production. As all HolosNor-models, the HolosNorDairy follows the Intergovernmental Panel on climate Change methodology. The output includes direct CH₄ emissions from enteric fermentation and manure management, direct and indirect N₂O emissions from manure management and soil, and CO₂ emissions from the production and use of farm inputs. In Norway, a collaborative initiative from the agriculture sector has resulted in the development of a greenhouse gas (GHG) calculator in the project of Climate Smart Agriculture. The primary objective has been to develop a practical tool for farmers and advisors to assess climate impact in agriculture and identify mitigation strategies at farm level. The initiative selected the HolosNor-models as they were tailored to production systems in Norway as the base for the calculator. The GHG calculator's unique feature involves gathering input data from various data sources, simplifying the process for farmers who only need to provide consent. Data sources for combined milk and meat production include herd information and energy requirements from the Norwegian milk herd recording system (Kukontrollen), invoices for concentrates, crop plans, manure planning, and milk and slaughter returns. The study aims to estimate GHG emissions for Norwegian combined milk and meat production using unique real-life data from the whole-farm GHG calculator. Although in early stages, the study will present GHG emissions for milk and meat based on approximately 800 unique farm estimations.

Assessing 'CO₂-warming-equivalent' emissions from Slovenian cattle production using GWP*

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Livestock farming, especially cattle farming, has been criticised for decades as a 'climate polluter', particularly because of its emissions of methane (CH₄) and nitrous oxide (N₂O), both of which are much more potent greenhouse gases (GHGs) than carbon dioxide (CO₂). However, GHGs with different lifetimes cannot be compared in terms of their impact on atmospheric temperature when calculated with internationally established GWP100 CO₂-equivalents (CO₂-eq). This is particularly important when comparing CH₄, which remains in the atmosphere for about a decade, with CO₂, which can remain in the atmosphere for a thousand years or more. Recently, new approaches have been developed to compare the warming effect of GHG emissions, considering their atmospheric lifetime. Here we calculate and compare emissions from Slovenian cattle production using CO₂-eq under GWP100 and 'warming-equivalents' (CO₂-w.e.) under GWP*. We used the IPCC and EMEP/EEA guidelines, adjusting Tier 2 to the Slovenian conditions, and the CO₂ equivalence from the Fifth Assessment Report (28 for CH₄ and 265 for N₂O). We found that the average GHG emissions from cattle production in the period 2018-2022 amounted to 1,245 kT CO₂-eq and 509 kT CO₂-w.e., with dairy cows averaging 458 kT CO₂-eq and -279 kT CO₂-w.e. and non-dairy cattle 787 kT CO₂-eq and 788 kT CO₂-w.e. Cattle production accounted for an average of 8.18% and 1.69% of all anthropogenic emissions when calculated using GWP100 and GWP*, respectively. As a result of past emission trends, the dairy cow sector expressed a 'cooling' effect, while the non-dairy cattle sector had a 'warming' effect. The trend of CO₂-w.e. was negative in all cattle categories. We conclude that the GWP100 approach overestimated the impact of Slovenian cattle production on climate.

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Public acceptance of microbiome management practices in dairy cows

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The aim of this study was to investigate public acceptance of innovative microbiome management practices aimed at improving the health and welfare of calves, as well as addressing environmental concerns. Data used in the analysis were obtained from an online survey conducted in July 2023. The final sample consists of 3 220 citizens from four EU countries (Finland, France, Ireland, and Poland). Participants were asked to evaluate the following interventions: (1) using colostrum to treat calves' diseases, (2) providing mixed probiotics powder, (3) administering probiotics through yogurt or kefir, (4) ensuring prolonged cow-calf contact, (5) supplementing feed with natural additives such as algae or seaweed extracts, (6) incorporating plant-based oils or fats, and (7) using the chemical feed additive 3-NOP to reduce methane emissions from dairy cows. Participants rated their acceptance of each practice on a standardized scale from 1 to 5, where 1 represented "strongly disagree" and 5 represented "strongly agree." The findings reveal that prolonged cow-calf contact was the most acceptable measure among tested interventions in all countries. Additionally, there was a significant difference in the acceptance levels between practices that use natural and chemical feed additives, with European citizens being more reluctant to use the latter. Interestingly, providing additional information to the participants resulted in a decrease in the acceptance of the measures, except for the use of chemical additives. Our findings have implications for encouraging broad acceptance of sustainable production schemes. The HoloRuminant project has received funding from European Union's Horizon 2020 research and innovation program under Grant Agreement No 101000213. This abstract reflects the views only of the author, and not the European Commission (EC)

Crossing grazing calendars and bulk milk analysis: is on-field validation for grass-diet in cattle possible?

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Managing an animal system based on pastures is a time-consuming activity that exhibits many benefits. Assessing the extent to which farms optimize pasture usage and prioritize grass feeding presents challenges. This assessment holds significance for various labelling requirements mandating a proportion of grass in the diet and serves to incentivize favourable management practices. Consequently, a verification method based on milk composition seems relevant. This study seeks to validate such hypothesis by cross-referencing 75 tank-milk analysis with 526 records from grazing calendars and supplementary feed data. Employing hierarchical clustering, distinct management practice groups were identified, with Groups 2 and 4 displaying the most discernible differences, particularly in their emphasis on full grazing and grass-based diets. Subsequent analysis revealed significant variations in milk composition among these groups, prompting the development of a predictive model based on Partial Least Squares-Discriminant Analysis (PLS-DA) from key milk traits predicted by milk mid-infrared spectrometry, like milk pH, Mg, K, Ca, P, C18:1 trans, C14:1 and C16. The study yielded interesting results, with an accuracy to distinguish Group 2 from Group 4 of 0.92. Using this PLS-DA indicator would be interesting as a next step to potentially determine the proportion of grass in diet and even the proportion of grazed grass. This could not only help in the verification for labelling, but also become an indicator for farmers to see if their cattle best valorise the pastures.

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Poster 13

Summarizing contributions dealing with environment and Green Deal

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The EU has high reaching goals: improve quality of water, strengthen the Nature 2000 areas, reduction of GHG emissions (by 90% in 2040 compared to 1990), create more biodiversity, and limit pesticide use (by 50% in 2030). This all is also part of the so called EU-Green Deal, that has been recently approved in Europe and, later on, came in debate again. This package of measures specifically affect the (intensive) agricultural and livestock regions. It has become a hot societal topic. In this specific contribution, the various contributions from speakers in this session and some other sessions will be discussed in a kind of round table set-up, stimulating maximum input from the audience. Views from different countries and continents may be helpful to grasp the various developments going on and strategies in place, dealing with environmental issues, and providing food for thought and further steps to take in research and in the field.

Trade-offs between biochemical methane potential of manure and enteric methane mitigation in dairy cows fed Bovaer®10 or Acacia mearnsii

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Batch incubations were conducted to evaluate how dietary supplements of 3-nitrooxypropanol (3-NOP; Bovaer®10) and tannin extract (TAN, Acacia mearnsii) affect the biomethane potential of dairy cow manure. Eight spot fecal and urine samples were collected from 16 lactating dairy cows fed 1 of 4 diets: basal TMR (CON), diets containing 3-NOP (60 mg/kg DM), TAN (3% of DM), and 3-NOP + TAN (4×4 LSD with 4, 24-d periods). The fecal and urine samples were composited by cow and period and combined into manure of a 70:30 fecal-to-urine ratio weighing 5.7±0.08 g and were prepared in quadruplicate (n = 252). Samples were suspended in 50 mL of H₂O and anaerobically incubated at 39°C (shaking at 100 rpm). Gas production (mL) and composition (gas-chromatography) were measured after 24h, 48h, and subsequently every week until CH₄ production decreased to <1% of the total cumulative production. Data were analyzed in R using mixed model. No dietary effect was observed for any of the cumulative gas production variables (P > 0.05) except the H₂ production, where TAN had a 2.27% lower (P < 0.05) production compared with CON. The average cumulative manure CH₄ across treatments was 6.96 mL. Initial NH₃ concentration and pH of fresh manure (1.45 mM and 8.17, respectively) were unaffected by diets (P > 0.05). Overall, there were no trade-offs between enteric CH₄ mitigation and manure CH₄ production. Further analysis will determine the organic matter content, and fermentation rate parameters in the manure and assess the overall impact of the tested enteric mitigation strategies on manure fermentation potential.

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Theatre 1

Lysine requirements of pigs divergent in breeding value for nitrogen efficiency

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This study aimed to relate the Lysine (Lys) content of the feed to the performance of pigs genetically divergent for nitrogen efficiency. From 40 litters over two batches, 120 three-way crossbred female grower-finisher pigs were grouped by high or low breeding value for nitrogen efficiency. In each group, pigs were assigned to three dietary treatments in a two-phase feeding program: low (8.4 g/kg Lys – 7.1 g/kg Lys), medium (9.0 g/kg Lys – 8.4 g/kg Lys), or high (9.5 g/kg Lys – 9.0 g/kg Lys) protein content. All diets had an equal mineral, vitamin, and energy level (9.59 MJ NE/kg). Pigs were offered feed ad libitum and individual feed intake was recorded. BW's were recorded at the start of the trial (37 ± 5 kg), before feed change (74 ± 7 kg), and at the end of the trial (121 ± 22 kg). ADG was 1040 ± 89 g/d during the first phase and 1154 ± 145 g/d during the second phase. ADG increased with 24 g/day for every extra g/kg Lys in the diet during the first feeding phase with no difference between the breeding value groups (P=0.760). ADG increased with 18 g/day for every extra g/kg Lys during the second feeding phase, and animals with a low breeding value for nitrogen efficiency grew on average 45 g/day less than animals with a high breeding value at similar Lys intake (P=0.017). The data did not show an optimum dietary Lys content for ADG, but rather indicated a linear increase in ADG with increased Lys intake beyond the levels tested in this experiment. Hence, this experiment showed that breeding for improved nitrogen efficiency will result in increased ADG at similar Lys intake and that the genetic potential for ADG might be higher than commercial levels of dietary Lys will attain. This study was part of the 3D'Omics project, funded from the EU's H2020 Program under grant agreement no 101000309.

Effect of dietary P depletion-repletion on pig bone characteristics

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The objective of the present study was to evaluate the influence of combinations of depletion-repletion periods of dietary phosphorus and calcium levels on bone characteristics in pig carcasses using computed tomography (CT). In this study, 48 entire male pigs were reared in pens (4 pigs/pen) and received two types of diets (C-control: recommended nutritional requirements; R-restriction: 60% of the nutritional requirements of digestible P and Ca) in three phases, resulting in 4 feeding strategies (control: CCC, deficit: RRR, compensatory 1: RCC and compensatory 2: RRC). At 24 h postmortem, 48 left carcasses were analyzed using CT (Philips Brilliance 16) and subsequently some bones were extracted (metacarpus III and IV and the 1st and 4th penultimate ribs) and mechanical tests were performed. The GLM procedure of SAS software was used considering strategy as fixed effect and pen as experimental unit. Tukey test was performed to find differences between strategies. The proportion of less dense bones (Hounsfield units +141 to +499) was higher in the RRR pigs than in other treatments ($P < 0.05$). Meanwhile, the proportions of bones of medium, high and very high density were higher in the CCC pigs than RRR pigs ($P < 0.05$), RCC and RRC treatments being in between ($P > 0.05$). On the other hand, in the mechanical properties of the bones analyzed, there were no significant differences between treatments. These results indicate that the conventional diet (CCC) could be substituted by the RRC or RCC strategies, in turn reducing dietary P with no effect on the bone characteristics and productive parameters during the growing of the pigs. Work financed by the project PID2021-124423OR-I00.

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Theatre 3

Impact of precision feeding during gestation on the productive career of sows over three reproductive parities

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This study aimed to evaluate the impact of precision feeding during gestation on the career performances and longevity of sows initially gilts, monitored over three reproductive cycles. Four isoenergetic treatments were compared: two conventional (CONV) constant-concentration feeding strategies (0.53% DIS Lys) with feed supply constant (flat feeding) or variable (bump feeding), and two precision feeding strategies (PF) based on the InraPorc model considering performance by parity or considering the individual weight of each sow at breeding. Four groups, representing 393 gilts, were followed from breeding to weaning over three cycles. The overall reproductive career was evaluated with a mixed model with contrasts opposing CONV strategies to PF strategies. Over the three reproductive cycles, the stillborn rate (-0.9 %; $P = 0.02$) and total piglet mortality (-1.5 %; $P = 0.03$) were lower in PF sows than in CONV sows. Overall, PF sows weaned more piglets during their three cycles than CONV sows did (+0.7; $P = 0.03$), with weaning litter weight gain also increasing for PF sows (+3.7 %; $P = 0.02$). The culling rate of gilts after three cycles did not differ significantly among treatments ($P = 0.27$) but tended to be higher for PF sows after one cycle (+4.9 %; $P = 0.08$). The results seem to show a lasting benefit of precision feeding for sows, with a decrease in piglet mortality during lactation, suggesting greater piglet viability without significantly decreasing sow longevity.

Impact of long-term precision feeding based on energy and lysine on the productive, economic and environmental performance of gestating sows

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Gestating sows are most often fed a single feed throughout gestation. This conventional feeding (CF) leads to deficits and excesses at individual level. Precision feeding (PF) is a way of addressing this problem, by adjusting the quantity and quality of feed for each sow on a daily basis. The aim of this study was to investigate the impact of a PF adjusted for energy and standardized ileal digestible (SID) lysine for gestating sows over 3 consecutive gestations. Quantities were adjusted using a 3-phase strategy over the gestation, with requirements estimated using a nutritional model for PF sows and farming feeding curves for CF sows. The PF sows received a diet which SID lysine content was adjusted daily to their needs; CF sows received a diet with a fixed SID lysine content. Trial began with 40 gestating sows allocated to a feeding strategy (PF or CF), based on their parity, body weight, and backfat thickness (BT). Linear mixed models were used to analyse the effect of the nutritional strategy over 3 gestations. The BT objectives at farrowing were achieved by PF sows over the 3 gestations, unlike CF sows who were below the objective at the 3rd farrowing ($P < 0.001$). Total SID lysine intake was reduced by 16% ($P < 0.001$), with decreased supplies in excess in the first 12 weeks of gestation thereby improving the efficiency of nitrogen by 4% ($P < 0.001$) for PF compared to CF sows. Applying a PF strategy resulted in savings of 10 € per ton of feed over 3 gestations without affecting the farrowing performances ($P > 0.10$). PF is a way of improving the sustainability of livestock farms by making a better use of the dietary nitrogen without compromising herd performance.

Effects of functional ingredients on intestinal health and growth performance of weaned piglets

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The aim of the present study was to evaluate effects of diets containing different functional ingredients on growth performance and gut health in post-weaning piglets. Six dietary treatments were tested using 720 male piglets (12 replicate pens per treatment) over a period of 34 d. The diets were ad lib fed during 13 d post weaning. It concerned a reference diet, the reference diet plus 3000 mg Zn/kg, as positive control (Control ZnO), a diet with 5% alfalfa meal (non-fermentable fibre), a diet with 5% sugar beet pulp (fermentable fibre), a diet with 3% BSF insect oil and a diet with 5% BSF insect chitin rich protein meal. During a subsequent three week period (d 13 – 34) all piglets received the same “phase II diet”. Transcriptomic responses in the ileal mucosa were assessed through RNA sequencing analysis conducted on samples collected at the end of both phases. In phase I ADG and ADFI were significantly higher and FCR improved ($p < 0.001$) in piglets fed Control ZnO compared to the other groups. However, in phase II and over d 0-34 performance did not differ between treatments. Piglets fed the Control ZnO diet recorded a better faecal consistency in phase I. Only in phase I, in Control ZnO piglets 39 genes were differentially expressed ($\text{adj_Pvalue} < 0.05$, $|\text{Log}_2\text{FC}| > 1$) compare to reference diet. The upregulated genes predominantly belonged to the metallothionein family (MT1A, MT3, MT1X), zinc transporters, and neurotransmitters (TH), while the downregulated genes were primarily associated with immune function (IL22, IL26, IL23R, IL17F, SFTPD) and oxidative stress response (NOX1). As evidenced by the outcomes observed in this study, in clinically healthy piglets, various potential functional ingredients related to gut health and function did neither improve performance nor gut functionality.

Nutraceutical Potential of Lentinula edodes' Spent Mushroom Substrate: A Comprehensive Study on Phenolic Composition, Antioxidant Activity, and Antibacterial Effects

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Lentinula edodes, or shiitake mushroom, is known for its potential health benefits. This work delves into the by-product of shiitake cultivation, spent mushroom substrate (SMS), to explore its applicability as a nutraceutical in animal diets. The SMS samples were collected and subjected to different extraction methods: short or long agitation and ultrasound-assisted extractions using different temperatures and distilled water or 50% (v/v) ethanol as solvents. The extracts were tested for phenolic content (total phenols, ortho-diphenols, and flavonoids), antioxidant capacity (DPPH, 2,2-diphenyl-1-picrylhydrazyl; ABTS, 2,2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid; and FRAP, ferric reducing antioxidant power), and antibacterial activity. The different extraction methods revealed significant variations ($p < 0.05$) in phenolic composition and antioxidant capacity. The highest phenolic content and antioxidant capacity were achieved using 24h extraction, agitation, 50°C, and ethanol as the solvent. Furthermore, the extracted compounds showed antibacterial activity in tested bacterial strains. This study highlights the nutraceutical potential of SMS, positioning it as a valuable dietary supplement for animal nutrition and emphasising its prebiotic properties. Acknowledgements: This work is supported by National Funds by FCT – Portuguese Foundation for Science and Technology, under the project UIDB/04033/2020 (<https://doi.org/10.54499/UIDB/04033/2020>).

Peptidomic profile of in vitro digested hemp-based products and their effect on intestinal epithelial cells

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In the last decade, attention in hemp-based products (*Cannabis sativa* L.) has increased. Among these, hemp-seed (HS) has gained interest from the feed and food sector due to high nutritional quality and bioactivity. In line of that, the aim of the present study was to determine the total phenolic content (TPC), antioxidant (ABTS, FRAP, ORAC) and ACE-inhibitory activity (ACE-IC) of in vitro digested HS and HS protein (HP), in relation to flaxseed (FX), largely used in the food/feed sector. In parallel, the peptidomic analysis and antioxidant effect of HS and HP on intestinal epithelial cells were investigated. The data (mean±SEM) were analysed by one-way ANOVA and considered statistically significant for a 95% confidence interval ($p < 0.05$). The results showed that the TPC of HP (1544.8±96.2mg Tannic acid equivalent (TAE)/100g) was significantly higher ($p < 0.05$) than HS (846.2±23.8mg TAE/100g) and FX (451.5±59.3mg TAE/100g). This trend was also confirmed for ACE-IC and ABTS (3936.8±59.3; 2386.6±117.9; 1334.5±43.6 mg Trolox equivalent (TE)/100 g, for HP, HS and FX, respectively). Most probably these positive effects are due to the bioactive peptides resulting from the hydrolysis of the main proteins with ACE-IC (eukaryotic translation initiation factor 6-2) and antioxidant (wall-associated receptor kinase-like 1) activity of HS and HP. These results encourage further investigation on the bioefficacy of HSs and HPs, to identify the best level of inclusion to ensure high performance and animal welfare.

Long-term effects of oregano essential oil supplementation in new born dairy calves

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We aimed to determine whether an early supplementation with EO in milk replacement improves short and long term growth, immunity microbiota, and metabolome in dairy calves. Sixteen dairy calves were randomized to two groups. The control and treated (EO) group (providing 0.23 ml of oregano EO for 45 days). After weaning, calves were kept in a feedlot and fed ad libitum with a ration formulated to cover their nutritional requirements. The animals were weighed, and blood and fecal samples were collected on days 3 (T0), day 45 (T1), and day 370 (T2). The EO group only had greater body weight and feed efficiency during the EO supplementation period. The CD14+ population average values were higher, whereas there was a lower abundance of Ruminococcaceae UCG-014, Faecalibacterium, Blautia, and Alloprevotella, and an increase of Allistipes and Akkermansia in the EO group. Some plasma metabolite changes, such as butyric acid, 3-indole- propionic acid, and succinic acid, particularly at T1, reflect parallel microbiota changes. Thus an early EO supplementation increased feed efficiency during the suckling period and modulated the dairy calves' immunological systems with notable changes in microbiota and plasma metabolome. No significant effects were evidenced later for metabolome and microbiome except for the large abundance of several primary metabolites.

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Theatre 9

Production efficiency during the early postpartum of dairy cattle supplemented with live *Saccharomyces cerevisiae boulardii* live yeast

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The transition period in dairy cows presents a complex phase, with both nutritional and physiological changes potentially affecting immune function and disease resistance. *Saccharomyces cerevisiae boulardii* has recently demonstrated health benefits in young ruminants. In a trial performed at the ILVO dairy cattle barn, 40 Holstein Friesian cows in transition (n=17 for parity 2 and n=23 for parity >2), received a control diet (CON) or a diet supplemented with *S. cerevisiae boulardii* CNCM I-1079 (1×10^{10} CFU/d) from days -28 till +35 from calving (SCB). Whereas no difference in total DMI postpartum was reported for cows in parity 2 (18.3 kg/d), a significant lower DMI was depicted in SCB cows in parity >2 compared to CON (19.2 vs 17.8 kg/d, P=0.012). The live yeast supplementation during early postpartum had beneficial effects on milk production, especially for dairy cows in their first transition period (34.7 vs 38.8 kg/d, P=0.014). Fat and protein corrected milk was also improved in parity 2. The nitrogen efficiency was higher in SCB cows, irrespective of parity (39.2 vs 41.1 %, P=0.02). Consequently, an improved feed efficiency independently of parity in SCB cows (2.04 vs 2.18, P=0.06) was depicted. NEFA and BHBA concentrations tended to be higher in plasma of SCB cows at start of lactation (Goossens et al., 2024), but stayed below the reported physiological reference values without leading to increased metabolic transition disorders. Total somatic cell counts was not different between group (4.6 log₁₀ SCC/mL). In conclusion, by promoting fat mobilization in a physiological range, *S. cerevisiae boulardii* can increase milk efficiency in early lactation without affecting the risk of metabolic transition problems.

Detecting microplastics in feces of pig fed former food products

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Former food products (FFPs) are authorized feedstuffs. However, there is presence of plastic packaging remnants in feed made from FFPs. Such contaminants, especially microplastics (MPs), can occur in biological matrices like feces. Thus, we aimed to test the efficiency of the selected method for MP sampling, digestion, and extraction in pig feces. The 36 fecal samples were collected from growing-finishing pigs fed a control diet or diets containing 30% sugary FFPs or 30% salty FFPs (n=12 per group). Feces were collected from rectum, placed in aluminum bags to avoid contamination, and 3 sets of 3 g feces were obtained from random spots in each fecal sample. Then, 25 mL 30% H₂O₂ was added to feces at room temperature for a week to digest organic matter. The 3 extra samples were spiked with blue polypropylene pieces to check if the digestion process may change plastic's appearance. For density separation, 100 mL saturated NaCl solution (D=1.2 g/cm³) was added to the samples that were filtered after overnight settling. Treated feces were inspected under a stereomicroscope to detect plastic remnants. Suspected particles were placed in a petri dish to be analyzed by the Fourier transform infrared spectroscopy for material confirmation. Plastic's appearance was not changed by H₂O₂ digestion. However, it was not able to detect any MPs from collected feces with the chosen method as the original packaging contamination in FFPs was low and MPs' presence was diluted by FFPs' inclusion level. Hence, further studies are needed to test different methods, develop a standard protocol for fecal MP extraction, and seek markers to trace the fate of MPs in animals.

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Effect of microplastic on rumen metabolism

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The environmental pollution by microplastics and their additives represents a problem for human health and food safety because they are accumulated in water, soil, and crops and transferred by animal feeding in milk, meat, and eggs. The role of bisphenols and phthalates as endocrine disruptors or carcinogens has been proved. Literature suggests that several bacterial species can metabolize microplastics or their additives. Hence, microbial communities of rumen could interact with microplastics and their additives resulting in a possible alteration of bacterial activities. This study aimed to evaluate the effect of several bisphenols and phthalates on rumen ecology using an in vitro trial. Bisphenols A, AF, F, S, C and monomethyl-, diethyl-, monobutyl-, dimethyl-, monobenzyl-, dioctyl-phthalates were individually fermented (0.83 mg/mL) using ewe rumen liquor as inoculum. Results showed that bisphenols affect differently microbial fatty acid metabolism compared to phthalates. The C18:0 (p<0.0001) concentration decreased while C18:2 c9c12 (p<0.0001), C18:2 c9t11 (p<0.0001), and C18:3 c9c12c15 (p<0.0001) content increased in fermenters containing bisphenols compared to the others. Monounsaturated fatty acid concentration was significantly affected by bisphenol presence in feeds (p<0.0001). The saturated fatty acid concentration was different between treatments (p<0.0001), with the highest value in the control fermenters and the lowest in the Bisphenol A ones. In conclusion, the additives of plastics interfered with microbial metabolism and the results on fatty acids showed a possible different action on microflora.

Dairy cows' diet discrimination through SIFT-MS-based volatolome of different matrices

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This study tested using SIFT-MS-based volatome of ruminal liquid, urine, faeces, sweat and milk to discriminate diets fed to dairy cows. Sixteen Holstein cows (1.38 ± 0.52 lactations, 74 ± 11 days in milk, 22.6 ± 2.74 kg kg/d of milk yield, 627 ± 44.2 kg LW, 1.4 ± 0.20 BCS, mean \pm SD) were enrolled in a longitudinal 10 weeks experiment based on two contrasting diets: 1) High-fibre and low-starch diet (HFibre, $n = 8$, 12 % starch DM basis), 2) High-starch (HStarch, $n = 8$, increasing starch until reaching 31 % in weeks 9 and 10). Cows were sampled at the end of weeks 9 and 10 and the samples stored at -80°C until the analysis. The volatolome was determined through selected-ion flow-tube mass spectrometry in full scan mode after thawing the samples at 37°C and heating them in water bath (30 min, 45°C). The data was analysed through partial-least square discriminant analysis for each sampling period independently. The number of ions ranged from 177 to 253 depending on the type of sample. While ruminal liquid, faeces and sweat did not yield predictive models at any time ($Q_2 \leq 0.21$ in all cases), urine yielded valid models with moderate-to-high predictive ability to discriminate the diet for both weeks 9 ($R_2 = 0.91$, $Q_2 = 0.74$) and 10 ($R_2 = 0.66$, $Q_2 = 0.51$). The milk volatolome also yielded valid models on both weeks 9 and 10, but its predictive ability was low ($Q_2 = 0.37$ and 0.47 for weeks 9 and 10, respectively). Our results suggest that urine, which is more related to the internal metabolism of the animal than the other matrices, has a better potential to be used for developing non-invasive techniques to follow the diet and nutritional status of productive animals.

Application of Faecal Near Infrared Spectroscopy to Measure Nutrient Utilisation in Horses, Cattle and Sheep

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Faecal near infrared spectroscopy has been implemented to monitor the nutritional status of animals in a non-invasive manner. Most calibrations developed are for single species, thus cannot be applied to predict dietary attributes in faeces of other animals. The objective of this study was to develop multispecies calibration models to predict faecal nutrient composition in herbivore species with varying digestive physiologies. Freshly voided equine ($n = 93$), bovine ($n = 89$), and ovine ($n = 80$) faeces were dried and milled and subsequently scanned in reflectance mode at 0.5 nm intervals in the visual-near infrared region of the electromagnetic spectrum, 400 nm to 2500 nm. Calibration equations were developed for individual species for each nutrient while multispecies calibrations were developed by combining samples from all three species ($n = 262$). Modified partial least squares regression was implemented during calibration development. Spectra were pre-processed using a combination of scatter corrections and derivative mathematical treatments. External validation was conducted to monitor calibration performance. Validation statistics for ash and organic matter (OM) for individual and multispecies models were satisfactory [ratio performance deviation (RPD) = 1.85 – 3.19]. Poor prediction accuracy was observed in the bovine model for dry matter (DM), neutral detergent fibre (NDF), and acid detergent fibre (ADF) [RPD = 0.95, 1.28 and 1.25]. Validation statistics for all nutrients predicted in equine faeces were very good, with excellent prediction of NDF [RPD = 5.47]. Calibration equations for all parameters predicted using the ovine model performed satisfactorily [RPD > 1.4]. Exceptional predictive ability was reported for crude protein using the equine and multispecies models [RPD = 5.22 and 6.01], followed by bovine and ovine models [RPD = 4.02 and 2.81]. All constituents were adequately predicted by the multispecies model, OM and ADF equations performed well [RPD = 2.78 and 2.45]. Overall, this study suggests that multispecies calibration models are suitable for the prediction of nutrients in faeces from animals with differing digestive physiologies.

A 7-Year Survey on corn silage nutrient composition in Po Valley (Italy)

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Corn is the primary crop in the plain of Northern Italy, a region characterized by intensive cropping and dairy farming activities. In particular corn silage is predominantly used as a base ingredient in dairy farms. Thus, monitoring the variability in the nutrient content of this primary ingredient is mandatory to meet animal requirements. Typically, drought-stressed corn silage is lower in energy overall (from 80–90% of a normal crop); 2022 was the hottest, driest year in Italy. Accordingly, the objective of this paper is to bring to light a significant volume of data on corn silage nutritional composition in Northern Italy. The Regional Breeders Association of Lombardy has been conducting a survey program to monitor corn silage nutritional composition over the last seven years (2017-2023). Nutritional composition was determined by NIRs while dry matter was also determined by oven drying. Mean values recorded over the experimental survey period were dry matter 34.6%, neutral detergent fibre (NDF) 45%, acid detergent fibre (ADF) 26.2%, acid detergent lignin (ADL) 2.7%, starch 29.1%, crude protein 7.3%, crude fat 2.4%, ash 3.9%, and forage unit (UF q.le) 29.6. Average values observed for nutritional composition were in line with literature results. However, considering the starch content, lower values were observed for the 2022 silage crop, furtherly numerous samples were below the 2nd quartile, indicating numerous samples with reduced content in starch. Concluding the present survey study provided information on the major effects of drought-stressed corn silage with a focus on a dairy dedicated area.

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Poster 15

Analysis of the sieve distribution of feces in dairy cows fed a starch-rich and a fiber-rich diet

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The assessment of feces is useful as a tool for control of cows. Differences in dry matter content or differences in the distribution of solids in the manure into coarse, medium and fine particles allow conclusions about the ration-composition and also about the digestive processes. The faeces of dairy cows from two feeding groups ([1] Starch and [2] Fiber) were used on 4 appointments. The faeces from 14 cows per feeding group were available (total 112 samples). The "RETSCH AS 200 BASIC" system was used for the present investigations. The manure samples were sieved using a RETSCH tower, and there were significant differences between the cows in the two feeding groups on the 4 mm sieve and the 1 mm sieve. It can be seen very clearly that the 4.5 % higher proportion of the fiber group on the 4 mm sieve is reduced by 5.2 % on the 1 mm sieve compared to the cows in the starch group. On the 1.25 µm sieve, no statistically reliable differences were found. In addition to the feeding group ($p = 0.001$), an effect of the sampling date ($p < 0.001$) were also determined with regard to the proportion of particles on the 4 mm sieve. The proportions on the 4-mm sieve were not influenced by the classification of the DMI of the cows with statistical certainty ($p = 0.080$), whereby the feeding group also had a significant influence on the proportions on the 4-mm sieve with $p = 0.007$. It could be observed that in both feeding groups the highest proportions on the 4 mm sieve can be detected with a high dry matter intake.

Discovery and functionality of bioactive peptides contained in the porcine intestinal mucosa hydrolysate product Palbio HP

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Porcine intestinal mucosa hydrolysates (PIMH) are a by-product of heparin production, obtained through a specific enzymatic hydrolysis which might generate bioactive peptides (BAPs). Our study aimed to characterize BAPs contained in Palbio HP (PHP, Bioiberica SAU, Spain), a PIMH protein source marketed for animal feed. Three PHP samples from batches 20/352, 20/0360 and 20/0361 were used. Each sample was fractionated based on molecular weight (<3 kDa, 3 to 10 kDa and >10kDa). The first fraction was directly analyzed by mass spectrometry (MS)-based peptidomics while the other two were first digested using LysC and trypsin and then analyzed by MS. The resulting peptide fragments were analyzed with MaxQuant and PEAKS software to identify and characterize peptide sequences. This workflow identified 961 peptides. Subsequent bioinformatic analysis using public databases (APD2, Neuropedia, StraPep, AHTPDB and BIOPEP-UWM) revealed the discovery of the following 6 significant BAPs and associated total amounts (pg peptide/□g sample): DAVEDLESVGK (0.1626), EGIPPDQQLIFAGK (0.2637), TITLEVEPSDTIENVK (0.3594), TNVPRASVPDGFSL (1.4596), TNVPRASVPDGFSLSEL (8.0500), VHVVPDQLMAF (0.0310). The first 3 BAPs are related to an antimicrobial activity, while the other 3 are associated with cytokine/growth factor-like, antioxidant or immunomodulatory activities. These activities are in accordance with prior in vivo benefits reported after using PHP in swine or poultry. In conclusion, PHP is a source of BAPs with biological functions which are consistent with its origin and preceding enzymatic hydrolysis process and could therefore potentially be used for enhancing animal health and welfare through dietary supplementation.

Impact of Precision Feeding and the “Bump Feeding” Strategy During Gestation on the Performance of Post-Weaning Pigs

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This study aimed to evaluate the post-weaning growth performance of piglets from sows fed with 4 isoenergetic treatments during gestation over 3 cycles: 2 conventional constant-concentration feeding strategies (0.53% DIS Lys) with feed supply constant (flat feeding; FF) or variable (bump feeding; BF), and 2 precision feeding strategies (InraPorc) considering the mean weight by parity (PFP) or the individual weight of each sow at breeding (PFI). At weaning, five litters from each of the dietary treatments were selected and from which five average piglets were transferred to a nursery building with 20 pens. All pens received the same feeding program in 3 phases of 14 days each. Growth performance was evaluated. Also, body lean and fat, and bone mineral content (BMC) of one piglet per pen were evaluated with the DXA scan (dual-X-ray absorptiometry). For growth performance, after phase 3, piglets from BF and PFI sows had a lower average daily gain (ADG) compared to FF, while PFP was intermediate ($P < 0.01$). The same was observed for the total post-weaning period ($P < 0.01$). Average daily feed intake was not impacted by the sows' treatment. For body composition, only the daily BMC gain from 14 to 42 days tended to be reduced for piglets from PFI sows compared to FF, PFP and BF being intermediate ($P = 0.07$). The daily lean and fat gain were not different between treatments. Following the finishing period of pigs from the same sows, dietary treatments during gestation did not impact their performance. Therefore, the feeding strategies used for gestating sows can influence the offspring's body composition and performance in nursery, with effects that seems more pronounced concerning energy intake.

Glutamate simultaneously increases the lean percentage and IMF content of Shaziling pigs probably via modulating the Spirochaetae-acetic acid axis

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This study aimed to explore the effect and mechanism of glutamate (Glu) supplementation on the growth performance, carcass traits, intramuscular fat (IMF) content, and gut microbiota of Shaziling pigs. Results showed that compared to the control group, Glu supplementation increased the average daily gain, final body weight, lean percentage, and IMF content, and decreased the fat percentage of Shaziling pigs ($P < 0.05$). In the muscle, Glu supplementation increased the concentrations of MUFAs, and decreased the content of C18:2n6, n6 PUFAs, and PUFAs ($P < 0.05$). Moreover, Glu supplementation significantly upregulated the mRNA expression of adipogenesis-related genes (FAS, SREBP-1C) and muscle growth-related genes (MyHCIIb, MyHCIIx), and increased the relative abundance of Spirochaetae and the acetic acid content in the colon ($P < 0.05$). Correlation analyses indicated that the acetic acid content was positively correlated with the relative Spirochaetae abundance and the IMF content, and tended to be negatively correlated with the fat percentage of Shaziling pigs. In conclusion, these results indicated that Glu supplementation could simultaneously increase the lean percentage and IMF content and decrease the fat percentage of Shaziling pigs, and these beneficial effects may be related to the Spirochaetae-acetic acid axis.

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Poster 19

Supplemental effect of dietary Almond hull on sows' and their offspring performance

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The cost for major traditional feed ingredients is continuously increasing for animal production, whereas utilization of low-cost by-products is not only economical but also sustainable. Studies reported that almond hulls as a fibrous resource have a medium nutritive value for swine. So, we intend to examine the effect of dietary almond hull (AH) supplementation on various aspects, including reproductive performance, nutrient digestibility, milk composition, and fecal score in sows and their progenies. From the 100th day of gestation to weaning d21, n=21 sows (Landrace × Yorkshire, parity of 3.3) were assigned to three dietary treatments: (i) control (CON), basal diet, and the basal diet supplemented with 3% and 6% AH. Reproductive performance, nutrient digestibility, milk composition, and fecal score data were analyzed using the GLM procedure of SAS, and the linear and quadratic test was used to determine the significance between treatment means ($P < 0.05$). Piglets born to sows fed AH supplementation showed higher ($P < 0.05$) body weight (BW) and average daily gain (ADG) at the end of weaning d 21 but the reproduction performance, and milk profile of sows did not exhibit any significant differences until end of the trial. In conclusion, the addition of AH supplementation to the sow diet would be beneficial to enhance the BW and ADG of their offspring without exerting any detrimental effects on the growth performance, nutrient digestibility, and milk composition in sows.

The use of near-infrared spectroscopy (NIRS) to predict feed value of Alfalfa (*Medicago sativa* L.)

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The aim of this study was to analyze the accuracy of near-infrared spectroscopy (NIRS) for simultaneous prediction and determination of several feed value parameters of Alfalfa (*Medicago sativa* L.): chemical composition and forage quality. A total of 451 Alfalfa samples representing a wide range in feed value parameters were scanned over the NIR spectral range (680 – 2500 nm) in reflectance, and the optical data were recorded as log 1/Reflectance (log 1/R). Meanwhile, actual feed value parameters: moisture, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), ash, total digestible nutrient (TDN) and relative feed value (RFV) of the Alfalfa samples were measured using proximate laboratory procedures. Calibration models were developed between chemical and NIRS spectral data using partial least squares (PLS) multivariate analysis in conjunction with spectral math treatments to reduce the effect of extraneous noise. The best calibration was selected based on the highest coefficient of determination in cross validation (R_2) and the lowest standard error of cross validation (SECV). The potential of NIRS to predict feed value of Alfalfa was the highest for Moisture ($R_2=0.96$, $SECV=5.75\%$), followed by ash ($R_2=0.87$, $SECV=0.38\%$), TDN ($R_2=0.77$, $SECV=1.50\%$), ADF ($R_2=0.77$, $SECV=1.92\%$), CP ($R_2=0.72$, $SECV=1.10\%$), RFV ($R_2=0.67$, $SECV=9.88\%$), and NDF ($R_2=0.52$, $SECV=2.55\%$). As a result, NIRS has demonstrated the potential to be utilized as a routine tool for estimating the main feed value parameters of Alfalfa.

Impact of Incorporating Hydrolysed Feather Meal on the Microbiota of Dogs Across Different Time Points

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This study aimed to investigate the effects of substituting 7% of poultry meat (CTR) with hydrolyzed feather meal (HFM) into the diets of female dogs on their microbiota composition after 15 days of adaptation (T0) and after 3, 7, 15 and 45 days (T3, T7, T15 and T45). Rarefaction curve analysis indicated optimal sequencing depth, evidenced by the asymptotic curve of the number of Amplicon Sequence Variants. Alpha diversity analysis revealed no significant differences between diets (CTR vs. HFM) and in relation to the Diet x Times interaction. Evenness analysis, reflecting both richness and relative abundance of taxa, showed higher values ($P<0.01$) in the HFM compared to the CTR diet. Diet x Time interaction depicted a higher and more homogeneous evenness value among subjects at time D0 of the CTR diet. The weighted unifrac distance matrix, related to the beta diversity, did not show significant difference between diets, with the CTR diet exhibiting a numerically higher value than the HFM diet. Our findings suggested that incorporating HFM into the diets of dogs have mild effects on their microbiota composition and that the hydrolyzed feather meal can safely substitute animal meat.

Diets rich in fiber during the transition period decreases sows' constipation prior to farrowing

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Constipation may result in partial occlusion of the birth canal and increased pain during farrowing, which can be associated with prolonged farrowing duration. Dietary fiber can be used to prevent constipation as they contribute to increased fecal moisture and faster transit time. The aim of this study was to evaluate the effects of a fibrous supplement in the transition diet of sows on fecal score around farrowing. Animals were randomly allocated at day 90 of gestation in one of the two groups: sows fed 2.6kg of control diet (CON; n=70; crude fiber=2.2%) and sows fed 2.8kg of fiber-supplemented diet (2.4kg gestation diet + 400g fibrous supplement, FIB; n=70; crude fiber=6%). Intestinal activity was monitored from two days before to two days after farrowing with the following score: 0= no feces, 1= dry and pellet-shaped, 2= dry and well formed, 3= normal and well formed, 4= normal/wet and formed, and 5= liquid feces. Variables were analyzed by logistic regression. Fecal score four and five were not observed. At two and one days prior to farrowing, the percentage of sows with fecal score 0 was greater ($p<0.05$) in CON-sows (CON = 35% and FIB = 7%; CON = 21% and FIB = 0%, for day 2 and 1 prior to farrowing, respectively). At the day of farrowing the percentage of sow with score 1 was greater ($p<0.05$) in CON sows (CON = 34% and FIB = 13%). FIB sows had greater proportion ($p<0.05$) of score 3 at two days before farrowing and at the day of farrowing. There was no difference on fecal score after farrowing ($p>0.05$). The results support the notion that CON-sows had a greater degree of constipation prior to farrowing. Increasing fiber content in the transition diet reduces constipation, and this may be translated in an easier farrowing process.

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Rumen content composition as an indicator of selective grazing patterns within three native South African sheep breeds

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Choosing livestock breeds that utilise resources more effectively and sustainably, can improve livestock production efficiency. This study aimed to identify selective grazing behaviour through differences in proximate rumen content composition of three South African sheep breeds. Dorper (n=12), Meatmaster (n=10) and Namaqua Afrikaner (n=13) castrates were kept on natural vegetation, without supplementation, on Nortier experimental farm, Western Cape, South Africa. Post-slaughter (156 days of age) a proximate analysis was performed on the dried rumen content. The rumen content of the breeds was compared using a one-way ANOVA. Namaqua Afrikaner castrates had higher rumen crude protein contents (12.17%) than Meatmaster (9.71%) or Dorper (7.58%) castrates. Namaqua Afrikaner rumen analysis showed higher crude fat ($p=0.012$), total digestible nutrients ($p=0.040$) and metabolizable energy ($p=0.040$) content than Dorper sheep. Dorper rumens had higher crude fibre percentages (48.8%) than the Namaqua Afrikaner (44.7%). Meatmaster castrates mainly had intermediate values. It seems that Namaqua Afrikaner rumen content had a higher nutritional value than the Dorper; suggesting that Namaqua Afrikaners select more nutritious shrubs than Dorpers. In continuous grazing systems this grazing pressure on nutritious shrubs may negatively affect biodiversity. Dorpers are less selective grazers that probably utilised a wider range of plants, having a lesser impact on the veld.

Silage Preservation Dynamics: Inoculant and Temperature Influence

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Efficacy for improved forage preservation as silage was assessed at 5°C, 20°C, and 35°C, with and without the addition of an inoculant, using 2.75 L PVC minisilos for 100 days. Whole-crop maize (cv. SY Glorius) with 44% dry matter (DM) was harvested at Research Farm Bottelare, Belgium, with a chop 6-8 mm length. Treatment with a silage additive containing *Lactiplantibacillus plantarum* IMI 507026 and *Pediococcus pentosaceus* IMI 507025 (Egalis® Ferment, Alltech Inc.) at 1×10^6 cfu/g was compared to a negative control (no inoculant). Seven replicates per treatment at 220 kg DM/m³ density were incubated at 5°C, 20°C, or 35°C. Lactic acid (LA) and acetic acid (AA) were determined by HPLC; and ethanol by NIR absorption. DM content at silo opening was measured by air drying at 60°C, corrected for volatile loss. DM loss, crude protein and ammonia were analysed, and NH₃-N,%TN was calculated as the ratio of ammonia nitrogen to total nitrogen. Data were collected after the ensiled period and analysed according to a Mixed Effect model where treatment and temperature were fixed factors and replicates random. Inoculant addition significantly ($p < 0.05$) reduced the DM losses, NH₃-N,%TN and AA concentration 48%, 15% and 12%, respectively compared to no inoculant group. Ensiling temperature significantly impacted the pH, LA, AA, ethanol and NH₃-N,%TN. These findings suggest that the temperature can influence the silage fermentation quality however, the inoculant addition can mitigate fermentation losses.

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Detoxification of deoxynivalenol and its derivatives by lactic acid bacteria in whole-crop maize silage

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Trichothecene concentration in whole-crop maize silage produced under different fermentation conditions (i.e. spontaneous or controlled via microbial inoculants addition) was examined in an ensiling trial using 2.75 l PVC minisilos. Whole-crop maize was harvested at the Research Farm Bottelare (Belgium) at a dry matter (DM) content of 44 %, and chop length of 6-8 cm. Treatment with a silage additive containing *Lactiplantibacillus plantarum* IMI 507026 and *Pediococcus pentosaceus* IMI 507025 (Egalis® Ferment, Alltech Inc.) at 1×10^6 cfu/g was compared to a negative control (no inoculant). Seven replicates per treatment were prepared at a density of 220 kg DM/m³ and incubated at either 20°C or 35°C. Analysis for the simultaneous presence of more than 54 mycotoxins was carried out using an LC-MS/MS method. Data were collected after an ensiled period of 100 days and analysed according to a Mixed Effect model where treatment and temperature were fixed factors and replicates random. Type B trichothecenes (deoxynivalenol (DON), 15-AcDon and 3-AcDon) were observed in most of the samples. DON was detected at the highest concentration, ranging between 0.45-2.68 mg/kg DM. Inoculant addition significantly ($p < 0.05$) reduced the DON by 74% (0.57 vs 2.22 mg/kg DM) and 15-AcDon by 38% (0.02 vs 0.05 mg/kg DM) compared to control, whereas the effect of temperature and the interaction were not significant. These findings suggest that Egalis® Ferment could contribute to the detoxification of DON and its derivatives in maize silage.

Effect of different Se-sources on milk biofortification: Performance and milk composition

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This study aimed to investigate the effects of different dietary Se-sources for milk biofortification on dry matter intake (DMI), milk yield and composition of dairy cows. Thirty-two mid-lactating Holstein cows (27.79 ± 6.29 kg milk/d, 2.06 ± 1.13 parities; and 128 ± 76.44 days in milk – DIM, at the start of the experiment) were enrolled into four treatments in a randomized block design: Control (no Se supplementation); Na-selenite [2.5 mg of Se as Na-selenite/ kg dry matter (DM)]; Se-yeast (2.5 mg of Se as Se-yeast/kg DM); and Se-methionine (2.5 mg of Se as Se-methionine/kg DM). After 28 days of supplementation, with 14 days for acclimation, cows were milked twice a day. Dry matter intake, milk yield, and composition were measured daily. All statistical analyses were performed in SAS version 9.4 (SAS Institute Inc.). Supplementation sources were analyzed and compared by using the following contrasts: C1 = CON vs. Se-sources; C2 = Na-selenite vs. Organic Se-sources; and C3 = Se-yeast vs. Se-methionine. Dry matter intake was increased for Se-sources compared to control treatment. In addition, Se-yeast treated cows had increased DMI compared to Se-methionine group. Despite the differences detected in the DMI variable, there is no effect of Se-sources supplementation on milk yield and composition. Overall, Se supplementation for biofortified milk production affected DMI, but did not alter milk yield and composition of mid-lactating Holstein cows.

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A multi-screening method based on ultra-high-performance liquid chromatography tandem high-resolution mass spectrometry (UHPLC-HRMS) to detect both regulated and emerging mycotoxins in different feed matrices

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Mycotoxins are secondary metabolites produced by filamentous fungi belonging to different genera, such as *Aspergillus*, *Penicillium*, *Fusarium*, and *Alternaria*. These compounds can cause several health damages for animals, thus determining reduced animal performance and profitability. In this work, we optimized a multi-screening method to detect both UE-regulated and emerging mycotoxins through UHPLC-HRMS Orbitrap platform, considering a resolution of 70,000 FWHM at 200 m/z. To test the overall performance, different feed samples including corn silages (CS), corn grains (CG) and grain mashes (CGM) (n = 31) collected from dairy farms located in Po Valley (Italy) were considered. We identified and accurately quantified 22 regulated and not-regulated mycotoxins, including aflatoxins (B1, B2, G1, G2; R₂:0.99, 1-50 mg/kg), fumonisins (B1, B2, B3, H-FB1, H-FB2; R₂:0.99, 10-100 mg/kg), trichothecenes (type A: HT-2 and T-2 toxins, R₂:0.99, 25-500 mg/kg; type B: DON, R₂:0.99, 100-1000 mg/kg), zearalenones (ZEA, α/β -zearalenol, α/β -zearalanol; R₂:0.99, 1-100 mg/kg), ochratoxin A (OTA; R₂:0.99, 1-500 mg/kg), alternariol derivatives (i.e., Alternariol, Alternariol monomethyl ether, tentoxin; R₂:0.99, 1-100 mg/kg), and tenuazonic acid (R₂:0.99, 10-250 mg/kg). Overall, fumonisins were the most abundant mycotoxins detected in both CS and CGM (0.19-1.06 and 0.29-0.39 mg/kg, respectively), while CG were mainly characterized by aflatoxins, being AFB1 in the range 0.76-1.10 mg/kg. The developed method will be tested in future applications based on emerging mycotoxins.

Effects of processing with Ozone on nutritional value of Grape pomace in sheep

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Circular feeding will benefit the world by reducing the carbon footprint associated with farm animal production and help in more sustainable production systems. Annual dried grape pomace (GP) production is estimating to five metric tons and it seems interesting to effectively circulate it into farm animal industry to mitigate environmental pollution. Lignin and condensed tannins limit GP nutritional value and we hypothesized that O₃ can be an effective, safe and green processing method to overcome these limitations. This study was aimed to investigate the effect of GP treating with O₃ in different time durations (6, 12 and 24 hours, n=5) and dietary inclusion of treated GP on performance of fat-tailed milking ewes. After in situ and in vitro evaluation, processing for 12 hours was selected and 48 ewes (n=12) were allocated to diets with 0, 20, 40, and 60 % of low quality alfalfa hay (CP 13.4, NDF 68.7, ADF 47.4, lignin 10.5, % of DM) replaced with GP. In terms of chemical composition, the average of laboratory replications (n=5) in each of experimental replication was employed for statistical analysis. For the in vitro fermentation, the average of intra- and inter-run replications within each experimental replication was used for statistical analysis with a CRD using CLM procedure of SAS, with significance threshold of $P \leq 0.05$. A regression analysis was done to determine linear or quadratic effects. The O₃ significantly reduced NDF (54.75 vs 42.85 % DM), ADF (41.57 vs 29.84 % DM), lignin (23.2 vs 12.3 % DM), total extractable tannins (6.48 vs 3.92 % DM) and condensed tannins (5.33 vs 2.19 % DM), but increased effective degradable NDF (29.29 vs 38.66 % NDF). Incremental dietary inclusion of GP didn't influence total VFA, but slightly reduced acetate concentration. The digestibility of NDF decreased but that of the CP and EE were not affected. Dry matter intake (2.38 vs 2.19) and milk yield (1.02 vs 1.11) was not significantly influenced, but milk fat percentage was decreased (5.42 vs 5.18) as a result of highest substitution rate. In conclusion, the processing of GP with O₃ improved its nutritional value and did not negatively influence lactational performance of ewes when replaced alfalfa hay.

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Effect of feeding hay or sugar beet pulp containing diets to gestating and lactating gilts on piglet plasma and ileum digesta metabolite concentrations after conventional and late weaning

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Feedstuffs rich in dietary fiber (DF) are part of welfare-friendly feeding concepts in pigs. This study investigated the effects of isoenergetic and isonitrogenous gestation (2.5–3.5 kg/day (d)) and lactation diets (ad libitum) containing 10% of a soluble (sugar beet pulp, SB) or an insoluble (hay, HA) fiber source ground fine (F, 1 mm) or coarse (C, 20 mm) (SB-F, SB-C, HA-F, HA-C) in 24 gilts (German Landrace, n=6/group). Whether DF in maternal diets affects metabolic profiles was tested in offspring (male, female) 2 weeks post-weaning (28 or 42 d) (2 piglets/gilt/weaning timepoint). Plasma was analysed by an enzymatic analyser and targeted metabolic profiling. Short chain fatty acids (SCFA) levels were determined in the ileum digesta using gas chromatography. Data were evaluated by ANOVA and Tukey test. Plasma concentrations of total protein, albumin, triglycerides, total cholesterol were higher while urea and free fatty acids were lower in late weaned piglets ($p < 0.05$). SB increased total cholesterol only while coarse particles increased total protein ($p < 0.05$). Metabolomics analysis identified 367 plasma metabolites with 54 differing between the treatment groups. Regardless of the particle size, SB and HA groups had 31 significantly different metabolites (amino acids, biogenic amines, and glycerol- and phospholipids), 25 of which were higher in SB than in HA. Ileum digesta concentrations of acetate, propionate, i-butyrate and total SCFA were increased in late weaned compared to conventionally weaned piglets in the SBP-F and SBP-C groups ($p \leq 0.05$), indicating an influence on the gut microbiota.

In vitro fermentation parameters of intensive beef fattening diets with sainfoin as hay or pellets in the concentrate

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Sainfoin (*Onobrychis viciifolia*) can be preserved as pellets or hay to be included in intensive beef cattle feeding systems, which typically use grain-based concentrates and straw in the Mediterranean area. An experiment was carried out to compare the in vitro fermentation parameters of three beef fattening diets recorded in an in vivo trial: 90% concentrate + 10% straw (C+straw), 80% concentrate + 20% sainfoin hay (C+SFhay), 90% concentrate including 15% sainfoin pellet + 10% straw (CSF+straw). Samples of the diets were evaluated in an in vitro assay using an Ankom system for 48 h in 3 separate runs. Gas and methane (CH₄) production, dry matter degradability (IVDMD), ammonia (NH₃-N), and volatile fatty acids (VFA) were studied. The diet affected gas and CH₄ production and the proportions of VFA ($P < 0.05$) but did not affect the CH₄:gas ratio, IVDMD, total VFA, and NH₃-N ($P > 0.05$). The diet CSF+straw produced more gas and CH₄ than the C+straw diet ($P < 0.05$) and more gas than the C+SFhay diet ($P < 0.01$), both with similar CH₄ production. Regarding the proportions of VFA, the C+straw diet had lower acetic acid and butyric acids and higher propionic, isobutyric, and valeric acids than the CSF+straw diet ($P < 0.001$); and only lower acetic acid and higher propionic acids than the C+SFhay diet ($P < 0.001$). In conclusion, compared to the C+straw diet, feeding sainfoin hay only modified the proportions of acetic and propionic acids, whereas the inclusion of sainfoin pellets in the concentrate had a greater effect on the in vitro fermentation parameters, increasing gas and methane production, and modifying most of the VFA proportions.

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Correlation with plasma leptin levels and body fat mass in mature beef cows grazing different herbage allowances of grasslands

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The aim of this study was to evaluate the effect of controlling the intensity of grassland grazing (Campos biome), through control of herbage allowance (HA), on distribution of body fat and plasma leptin levels in beef cows of different genotype. Mature beef cows ($n=32$) were used in a complete randomized block design with a factorial arrangement of herbage allowance (2.5 vs. 4kgDM/d; LO vs. HI) of native pastures (52%DM, 8.4%CP, 39.7%FDA,) and cow genotype (CG; Angus- Hereford vs. F1 reciprocal crosses; PU vs. CR). The experiment was conducted for 3 years and at the end of the third year at 192 ± 10 days postpartum cows were slaughtered and weight and samples of all tissues were collected for chemical composition analyses. Prior to slaughter, cow BW and BCS were recorded, and blood samples were collected. Means from a mixed model analysis were considered to differ when $P \leq 0.05$. Slaughter BW tended ($P=0.08$) to be greater for CR than PU cows. Body fat mass (BFM) or carcass fat and BCS were not affected by HA, CG or their interaction. The visceral fat mass was affected ($P=0.05$) by the interaction between HA treatment and CG. Plasma leptin levels was not affected by HA, CG or their interaction. Plasma leptin levels was positive correlated with: BW ($P=0.03$, $r=0.42$), BCS ($P=0.03$, $r=0.43$), BFM ($P \leq 0.01$, $r=0.78$), carcass fat ($P \leq 0.01$, $r=0.80$). Though there was no effect of HA treatment or CG on the quantity of body fat mass and plasma leptin level, the animals with more BFM are those who had higher levels of plasma leptin. The high correlation between plasma leptin levels and fat carcass that is consistent with this type of fat that synthesizes this hormone.

Water Quality: Farmers' perception of the importance of the quantity and quality of water used for dairy cattle in Portugal

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Climate change affects livestock systems through direct impacts on the physiology, behavior, production and welfare of animals and indirectly through the availability, composition and quality of food and milk. The aim of this study is to ascertain whether dairy cattle farmers in Portugal (1) consider the issue of "water scarcity and quality" to be important on dairy cattle farms. Questionnaires were sent to dairy cattle farms in mainland Portugal. The farms (n=285) were selected based on their location, using the databases of the Portuguese Association of Friesian and the Dairy Cattle Support Station. The questionnaires aimed to collect information on (1) the importance of water attributed by producers; (2) water monitoring and storage; (3) production, reproduction and animal health; (4) verification of the latest analysis water report. The preliminary results of the questionnaires indicate that 51% of farms consider that water quantity alone is the most important factor on dairy farms; 98% of producers say that, in the context of climate change, water scarcity is very worrying; 91% of farms use their own water (borehole); 40% of farms do NOT carry out water quality analyses; 86% of farms do NOT monitor water consumption; 88% of farms do NOT carry out any kind of water treatment. The main problem found in the water analyses consulted was associated with microbiological quality (presence of *E. Coli* >0 NMP/100ml; intestinal enterococci > 0 cfu/100ml), followed by the presence of iron (>3mg/L), nitrates (>200 mg/L) and manganese (>4mg/L).

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Poster 33

Effect of pastures with rich tannin legumes on milk production and beef calves' performance

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The aim of this study was to evaluate cow's milk production (MP), and beef calves' performance grazing native improved pastures with a rich tannin legume (*Lotus subbiflorus*). Fifty-six Aberdeen Angus×Hereford multiparous suckling cows (LW 497±6.8 kg; BCS 4.05±0.04 units; Calf LW 60±1.6 kg) were assigned to each of two treatments on Day 22±0.82 postpartum: (i) native pasture (NP; n=28); (ii) native pastures improved with *Lotus subbiflorus* (improved pastures, IP, n=28). The forage allowance was 4.7 kg DM/kg LW in both treatments. The proportion of legume in IP was 27% with 3.6g/100 g DM of condensed tannins. Milk yield was assessed between onset of the experiment until Day 133 postpartum biweekly using a milking machine after oxytocin injection. The data were analyzed with the PROC MIXED of SAS (9.0; USA) with a model that included the fix effect of treatments. Cows LW was higher (P<0.05) in IP treatment respect to NP on Day 78 (546 vs. 523 kg, SEM=7,6) but not on Day 133 postpartum (551 vs. 539 kg for IP and NP respectively; SEM=7,8). Average MP (kg/d) was higher in IP than NP cows from Day 30 to 84 postpartum (12.3 vs 10.8, SEM=0,24; P<0,005) and from Day 84 to Day 140 postpartum (8.8 vs 7.7, SEM=0.31; P<0.05). Calves DLW between 22-78 days postpartum tended to be higher (P=0.057) in IP than in NP (1.13 vs 1.07 kg/d, SEM=0.02) but no differences were observed between 78-133 days postpartum between groups (1.08 vs 1.09 kg/d, SEM=0.24, for IP and NP respectively). Under grazing conditions, the incorporation of *Lotus subbiflorus* on native pastures is a nutritional and economical management strategy to increase postpartum beef herd performance.

Apparent digestibility of cull chickpeas and whole grain of corn meal in growing pigs

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The objective of this research was to determine the influence of soybean meal and whole grain of corn meal for cull chickpeas on apparent digestibility of nutrients in growing pigs; six crossbred pigs (Average body weight 50.62 kg) were used in a replicated Latin Square Design. Pigs were assigned to consume one of three diets: 1) Diet with 17.99 % CP and 3.12 Mcal ME/kg, containing whole grain of corn meal 70.0 %, soybean meal 27.5 %, and premix 2.5 % (CONT); 2) Diet with 17.52 % CP and 3.17 Mcal ME/kg with 56.5 % whole grain of corn meal, cull chickpeas 20 %, soybean meal 19 %, vegetable oil 2 %, and premix 2.5 % (CHP20), and 3) Diet with 17.50 % CP and 3.17 Mcal ME/kg containing whole grain of corn meal 43.0 %, cull chickpeas 40 %, soybean meal 11.5 %, vegetable oil 3 %, and premix 2.5 % (CHP40). Pigs were individually placed in metabolic crates (0.6 × 1.2 m). The adaptation period was 6 days and sample collection period was 4 days. From each diet and period, one kg of diet was taken as a sample and the total fecal production was collected. Apparent digestibility of DM with values of 80.19, 81.05 and 79.95 %, for CONT, CHP20, and CHP40, was not affected among treatments (P>0.05). Apparent digestibility of crude protein was similar (P>0.05) by CHP20 and CHP40 diets (79.15, 78.95 and 78.60 %), and apparent digestibility of OM was not affected (P>0.05) by CHP20 and CHP40 inclusion (82.45, 81.55 and 80.75 %). It's concluded that cull chickpeas can be used in growing pig improving nutrient digestibility, by comparing soybean-whole grain of corn meal diets.

Leucine regulates lipid deposition by promoting the secretion of adipokines and the SIRT1-PPAR γ pathway

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This study aimed to investigate the molecular mechanism of leucine (Leu) on the regulation of lipid deposition in adipocytes. 3T3-L1 preadipocytes were taken for directional induction differentiation, and 0.5 mM, 1 mM and 2 mM Leu were added to the medium of the treatment groups for 8 days, until the control group was completely differentiated, and each test was repeated 3 times. All data were analyzed by ANOVA using the IBM SPSS 26, regarded as statistically significant at P < 0.05. The results showed that: With the increase of Leu concentration, 1) the number of lipid droplets and the content of triglycerides in adipocytes decreased, and the content of glycerol and free fatty acids in the culture supernatant increased; 2) The levels of leptin, resistin, RBP4, TNF α and IL-6 in the adipocyte culture supernatant decreased, while the level of adiponectin increased (P < 0.05); 3) After Mito-Tracker fluorescence staining, the number of lipid droplets decreased and the mitochondrial fluorescence intensity increased, while the relative copy number of mitochondrial DNA of adipocytes increased (P < 0.05); 4) Leu down-regulated the mRNA expression level of lipid metabolism-related genes such as PPAR γ and PLIN, up-regulated the mRNA expression levels of mitochondrial function-related genes such as HSL and PGC-1 α and Tfam, and increased the protein expression levels of P-mTOR, P-S6K1, P-4EBP1, P-AMPK, P-SIRT1, PGC-1 α , and PKA, while the protein expression levels of PPAR γ were decreased (P < 0.05). 5) The expression of intracellular lipid deposition marker protein PPAR γ was down-regulated by the construction of SIRT1-overexpressing adipocyte lines. Therefore, Leu can reduce lipid deposition in adipocytes by affecting SIRT1-PPAR γ signaling pathway and mTOR signaling pathway, regulating adipokines' secretion and increasing mitochondrial biosynthesis, which may be a potential nutrient regulatory strategy and target for increasing lean meat rate, improving feed conversion rate and alleviating nutritional metabolic diseases in pig productivity.

Dairy and plant-based proteins as FBS alternative in muscle cell cultivation

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Abstract The use of Fetal bovine serum (FBS) in mammalian cell culture due to its complexity, ethical issues, and batch-to-batch variability raises safety concerns for applications as cultivated meat and milk production. Here, we showed two sustainable alternatives, whey proteins (WP) and hemp seed proteins (HP) to support C2C12 muscle cell growth. Accordingly, cells exposed to FBS (positive control, 10-0%), individual WP (10-0%) or HP (1-0 mg/mL) in DMEM for 48 h (day 1, and 2) to proliferate. Subsequently, the treatments were replaced with 2% horse serum medium until full differentiation (day 6). WPs included α -lactalbumin (LA), β -lactoglobulin (LG), bovine serum albumin (BSA) and lactoferrin (LF). Cell viability and lactate dehydrogenase (LDH) release accessed on day 1, 2, and 6 showed LA and LG to maintain viability until 5%, BSA 2.5%, and LF 0.62% compared to 0% ($p < 0.05$, ANOVA). Above this range inhibited viability or enhanced LDH release ($p < 0.05$). HP maintained viability at all concentrations tested. From dose-reponse study, WP mixtures as high (LA 1.25%, LG 1.25%, BSA 1.25%, LF 0.6%) and low (LA 0.15%, LG 0.07%, BSA 0.15%, LF 0.03%) were prepared and cell proliferation and differentiation assessed. Low WP mixture promoted cell growth as FBS while, exceeded other treatments groups. Also, WP and HP efficiently induced differentiation as FBS. This study shows WPs and HP as promising alternatives for cultivated food production. Project funded-NRRP, Mission 4 Component 2 Investment 1.3-Call for tender No.341, 15 March 2022, Italian Ministry of University and Research funded by EU – NextGenerationEU; OnFoods, Project code PE00000003&Prin2022.CELL to FOOD²-G53D23003940006

NX pig-derived lactobacillus modulates host intramuscular fat deposition via branched-chain amino acid metabolism

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Gut microbiota has been extensively demonstrated to modulate host lipid metabolism. Higher intramuscular fat (IMF) accumulation of Chinese indigenous breed pigs is associated with their special gut microbiota structure. However, the specific microbes and metabolic pathways responsible for lipid deposition are still not well understood. In the present study, a comparative analysis of the gut microbiota and metabolome in obese NX pigs and lean DLY pigs was conducted. The results revealed a higher abundance of gut lactobacilli and a correlation of branched-chain amino acid (BCAA) metabolism pathway in NX pigs. We proceeded to verify the roles of various lactobacilli strains originating from NX pigs in BCAA metabolism and lipids deposition in SD rats. We demonstrated that *L.reuteri* is a fundamental species responsible for modulating lipid deposition in NX pigs, and that increased circulating levels of BCAA are positively linked to greater lipid deposition. Additionally, it has been verified that *L.reuteri* originated from NX pigs has the ability to improve BCAA synthesis in the gut and enhance IMF content in lean DLY pigs. The expression of genes related to lipid synthesis was also significantly upregulated. Collectively, our results imply that NX pig-derived *L.reuteri* regulate BCAA metabolism and thus has a potential role in improving meat quality of lean pig breeds through modulation of host intramuscular lipid deposition. The study's findings present a new approach to altering gut microbiota composition, enhancing the quality of meat, and exposing the part of gut microbiota-mediated BCAA metabolism in organismal lipid metabolism.

The use of Maillard Reaction Products and reduced CP in pig finishing diets

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This study investigated the effect of a low protein diet integrated with a Maillard reaction product (MRP) on performance, water consumption, NH₃ production and slurry production. Three groups of about 600 pigs were reared from 16 to 140 kg. The control group (C) used three rations with crude protein (CP) of 16/15.3/13.9%. The second group (LP) used a lower CP (16/14.6/13.5) in the 2^o and 3^o diet with the same AA profile and energy concentrations. The third group (DG) used a lower CP diet (16/14.6/13.5) in the 2^o and 3^o diet integrated with a MRP, the same AA profile and energy concentration. There was no significant difference in production performance between the groups with an average daily gain of 845 g and a food conversion of 2.6. The water consumption in the LP and DG groups was significantly reduced ($p < 0.001$) using a one-way ANOVA analysis with Day as a blocking factor. The DG group tended ($p < 0.08$) to have a lower NH₃ production using a Regression analysis due to unbalanced design, using Day as a blocking factor. The DG group produced a total of 23% less slurry compared to the C group. The use of specific Maillard Reaction Products which bonds lactose, fructose and sucrose to lysine, methionine and threonine, enable a constant supply of AA release after digestion which permits the use of a lower CP diet. This study was carried out in one of the most pig dense areas of Europe (Llerida, Spain) where NH₃ production and slurry disposal is a major problem. MRP products will permit the use of lower CP diets and lower the consumption of imported soya bean meal and the carbon footprint of pig production. Water is tending to be a limiting factor in some areas of Spain which has the highest pig production in Europe and this study clearly shows savings in water consumption.

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Theatre 1

Decreasing feed-food competition by use of cereal by-products in grower-finisher pig diets

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Pig diets typically contain about 60% cereals and 10-15% SBM. To decrease feed-food competition, partial or total replacement of cereal grains by cereal by-products (BP) in the diet of growing pigs was studied. The influence of BP inclusion on growth performance, carcass characteristics and environmental impact was examined with or without reduction in dietary energy to allow more BP inclusion. A completely randomized block design was used with 8 treatments in a 2x4 factorial arrangement with dietary energy level (9.5 or 10.0 MJ NE/kg) and replacement of SBM, palm kernel meal and wheat (0, 33, 67 and 100%) by BP (maize DDGS, wheat gluten feed meal and wheat middlings) as respective factors. In total, 14-16 replicate pens with 6-10 Duroc*TN70 male or female pigs from 23-125 kg, were included per treatment. Body weight, ADG, feed intake, FCR, faecal consistency, N and P efficiency and dietary carbon footprint (CFP) were analysed using a generalized linear mixed model and pen as experimental unit. Overall, ADG was 1004 g/d. Increasing BP reduced ADG ($P < 0.001$) by up to 100 g/d, irrespective of dietary energy content, largely due to a reduction in feed intake ($P < 0.001$) of 140-200 g/d. Increasing energy level enhanced ADG by 25 g/d while reducing feed intake by 45 g/d ($P < 0.05$). Overall, highest inclusion of BP impaired FCR by 3%, with more effect at low than high dietary energy content ($P < 0.05$). Increasing dietary BP reduced carcass dressing percentage, increased backfat and reduced muscle thickness and lean meat% ($P < 0.01$). Moreover, increasing BP did not consistently affect N efficiency, but substantially reduced P efficiency by up to 16 %-units ($P < 0.001$) due to a high P content of cereal BP. Overall, increasing BP reduced CFP per kg carcass gain including land use change (LUC) up to 16% ($P < 0.001$) but increased CFP excluding LUC ($P < 0.001$) by up to 27%. In conclusion, increased use of cereal by-products to reduce feed-food competition and improve circularity of pig production may impair growth performance and other sustainability criteria. Adequate insight in the nutritive value of the BP and their impact on performance characteristics is required to minimize these trade-offs.

Cereal by-products in diets for weaned piglets

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To enhance sustainability of pig production, the concept of circularity has gained increasing attention. Circularity can be improved by replacing human-edible feed ingredients as cereal grains by alternative ingredients as by-products and food unfit for human consumption. The aim of this study was to determine the effect of total or partial replacement of wheat by cereal by-products (DDGS, maize feed meal and wheat gluten feed meal) in the diet of weaned piglets. In total 660 piglets (weaning age 27 days) were allocated to 4 dietary treatments at D7 post-weaning. Treatments were: (1) control diet; 39% wheat; and diets with (2) 33%; (3) 67%; (4) 100% replacement of wheat with cereal by-products. The diets were fed from D7-D28 post-weaning. Data were analysed with ANOVA using Genstat statistical software with pen as the experimental unit. Replacement of wheat tended ($P=0.06$) to linearly reduce daily gain and linearly ($P<0.01$) increased feed conversion ratio. Inclusion of cereal by-products resulted in a significant linear decrease in digestibility of organic matter, and crude protein, and a linear increase in digestibility of crude fat and NSP. Replacing wheat by cereal by-products significantly reduced the N and P-efficiency and increased the carbon footprint of the diets by 6.2%, i.e. from 865 to 919 g CO₂-eq including LUC per kg of feed. These results indicate that complete replacement of wheat by cereal by-products as used in this study negatively affects carbon footprint, performance, and efficiency of N- and P-utilisation of the pigs. These effects are largely caused by the lower digestibility and high total N and P content of the co-products.

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Effect of sire type and a by-products based diet on performance and meat quality in growing-finishing pigs

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There is an increasing interest in using by-products in pig feeding. However, it's unclear whether the past selection for fast and lean growth has affected the capacity to cope with by-products. A 2×2 experiment was set up to investigate how crossbreds, sired by boars differing in feed intake, cope with alternative ingredients. Boars were from the same Piétrain line with either a high (HFI) or low (LFI) estimated breeding value for daily feed intake (DFI; difference of 3 genetic standard deviations). Between 14 weeks of age and slaughter, a cereal and soybean-based control (CON) diet was compared to a by-product-based diet high in fat and fiber (HFF). HFF diet was formulated to contain the same net energy, crude protein and digestible amino acid levels without any cereals or soybean meal. In total, 48 pigs per sire type × diet were included in the experiment. Performance, carcass and meat quality were measured. None of the traits showed an interaction ($p<.05$) between type of boar and diet. HFI pigs showed higher DFI and daily gain (DG) ($p<.001$), with no difference in feed conversion ratio (FCR) ($p=.205$), lower carcass quality ($p<.001$) and higher intramuscular fat ($p=.030$). For both boar types, pigs fed the CON performed better, with a higher DG ($p=.028$), DFI ($p=.011$) and dressing yield ($p=.009$) but without differences in FCR or meat quality. In conclusion, different types of pigs may cope equally well with diets high in by-products.

The impact of white sorghum inclusion in growing-finishing pig diets on growth performance and feed digestibility

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This study evaluated the impact of dietary replacing of maize, wheat, and soybean meal by a white sorghum (WS) variety, with high productivity, nutritional value (14% CP) and adaptable to Mediterranean drought conditions, on growth performance and feed digestibility in growing-finishing pigs. A total of 528 pigs ([Landrace x Yorkshire] x Pietrain), weighing $31 \pm \text{SD } 4.5$ kg, were distributed across 48 pens based on weight and sex. The study included three phases: Growing 1 (G1; d0-29), Growing 2 (G2; d29-46), and Finishing (F; d46-95), each corresponding to different feed formulations. Two dietary groups were established: one with conventional ingredients (maize, wheat and soybean meal; CON), and the other completely replacing maize and partially replacing wheat and soybean meal with a 50% inclusion of WS (SOR). Growth performance and carcass characteristics at slaughter were analyzed. One composite fecal sample per pen was collected at the end of each phase (d26, d42 and d84) for ATTD digestibility analysis. Data was analyzed using linear models, using the pen as the experimental unit. Pigs fed the SOR diet exhibited lower average daily gain (ADG) across the entire study (CON: 794 g/d; SOR: 776 g/d; $P = 0.034$). No effects were observed for BW (CON: 106.3 kg; SOR: 104.9 kg) and feed-to-gain ratio (FGR; CON: 2.1; SOR: 2.13). Pigs fed the SOR diet displayed lower ADG (CON: 828 g/d; SOR: 782 g/d) and higher FGR (CON: 1.76; SOR: 1.83) in G1, but there were no differences in G2 and F. Additionally, carcass yield (CON, 79.6%; SOR, 79.3%) and lean percentage (CON, 64.3%; SOR, 64.1%) were not affected by WS inclusion. Lower gross energy ATTD was observed for SOR diet in G1 (CON: 84.7%; SOR: 81.1%) and G2 (CON: 89.8%; SOR: 85.6%; $P < 0.05$) but not in F phase (CON: 90.7%; SOR: 90.1%; $P > 0.1$). In conclusion, replacing maize, wheat, and soybean meal by a 50% inclusion of white sorghum (14% CP) may become suitable alternative for finishing diets, promoting the use of local feed ingredients in swine diets.

Effect of two phases olive cake on diet digestibility and parameters productive in Bísara breed pigs

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The aim of the present study was to investigate the effects of two phases olive cake (OC) on the apparent total tract digestibility (ATTD) of the diet and dry matter intake (DMI) in 12 Bísara pigs (males castrated and females, 91.63 ± 3.29 initial BW, growing-finishing). The animals were distributed among 3 diets with 0, 15 and 25% of OC inclusion (OC0, OC15 and OC25, respectively), under ad libitum feeding, percentages of 17.2, 23.8, and 36.6% fiber, respectively. In this study, the digestibility of dry matter (DM), organic matter (OM), crude protein (CP), crude fat (CF), and fiber (NDF) were evaluated. In this study, the digestibility of dry matter (DM), organic matter (OM), crude protein (CP), crude fat (CF), and fiber (NDF) were evaluated (total feces collection, 4 days). In animals that received the diet with CO incorporation, the average daily intake (ADI) of CF (90g/day to approximately 170g/day) and NDF (886 to 1115g, in the CO25 group) increased. Incorporating 25% of OC reduced the apparent total tract digestibility (ATTD) of various parameters relative to the CD group. The OC15 group is similar to CD and OC25, except for the ATTD of CF, which is only similar to the CD group. Digestible DM, OM, and CP intake were not affected ($P > 0.05$), however, CF intake was higher ($P = 0.002$) in the OC-receiving groups (82.54 to 153.78g) than in the CD group (140.0g). Regarding the entire period, ADG and feed conversion ratio were not significantly different ($P > 0.05$). ADI was lower in the CD group (2.84kg) and increased in animals receiving OC15 and OC25 incorporation (3.46kg and 3.64kg), respectively. Final BW showed a tendency ($P = 0.056$) to increase in the OC15 group. In this exploratory trial, it was observed that the reduction in digestibility in animals receiving OC diets was compensated by an increase in intake. This pattern is consistent with results from previous studies, potentially being attributed to natural variation among the animals. Acknowledgments: This work was supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Effects of peat and lignocellulose on the colonic fermentation of pigs in an ex vivo simulation model

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Peat is a natural source of fiber and humic acids, both known for beneficial effects towards gut microbiota. Lignocellulose is frequently used fiber in pig diets. The present study compared peat and lignocellulose on the colonic microbial fermentation of pigs in an ex vivo simulation model. Before the simulation, the materials were treated with pepsin-HCl and then with bile acid, pancreatin and NaOH, to mimic pre-colonic digestion. Simulation treatments were 1.) control (no amendments), 2.) lignocellulose, and 3.-5.) peat from three locations A, B and C (all by Neova Oy, Finland), and they were tested at 1% concentration and with 4 replicates. The authentic growth medium was prepared from the distal ileal, cecal, and proximal colonic contents of two pigs of ~30 kg. Microbial inoculum was collected from the distal colon of one pig. A 16-hour simulation was performed under anaerobic conditions in closed vials with a volume of 15 ml at 37°C, using a gyratory shaker for mixing. Samples were collected at 6 and 16 hours using a needle and syringe, and analyzed for gas volume, pH, and short chain fatty acid profile. Data was analyzed by ANOVA and Tukey's test. Lignocellulose had no effects on the measured parameters, compared to control. Peats B and C significantly decreased pH at both time points. At 16 hours, peats B and C showed less branched chain fatty acids (BCFAs), which are considered to reflect fermentation of amino acids. Results of pH and BCFAs for peat A did not differ from control or from peats B and C. In conclusion, peat as a fiber source may promote beneficial colonic fermentation in pigs, potentially offering advantages over lignocellulose. However, further verification of the results through in vivo studies is needed.

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Partial or complete replacement of soybean meal with local protein sources in the diet of slow-growing broilers

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Broiler feed production in Europe is highly dependent on imported soybean meal (SBM) as the primary protein source, posing environmental concerns. Local soybean production is currently uncompetitive, but there are promising alternative protein sources in Europe. Besides legumes and oilseeds, black soldier fly larvae meal (BSFM) and processed animal proteins (PAPs) obtained as by-products from pig slaughterhouses are available for poultry after the recent changes in legislation. Transition to a complete supply of fresh chicken meat from slow-growing broilers (SG) by Dutch supermarkets emerged in 2021 to address consumers' animal welfare concerns, however, little is known about the impacts of replacing imported SBM with circular protein sources in diets of SG. This study aimed to evaluate partial or complete SBM replacement with combinations of different local alternative protein sources, i.e. BSFM, PAPs, field beans, peas, sunflower seed and rapeseed meals, on the production performance of SG. This study consisted of 6 treatments and 8 replicates and lasted for 55 days. Day-old male Hubbard JA757 broilers were assigned to 48 floor pens (19 birds/pen). From day one, birds were fed either a control or one of the 5 experimental diet programs that were isocaloric and contained the same digestible content of essential amino acids at each feeding phase. In general, partial or complete replacement of SBM with plant-based protein sources and BSFM did not affect body weight gain. Replacing SBM with plant-based proteins and PAPs negatively affected body weight gain (-4.2%) and feed efficiency (-4.4%) necessitating further analysis of the PAPs. Replacing SBM with BSFM and plant-based proteins resulted in the highest body weight gain until 24 d (+4.0%) and the best feed efficiency among treatments, but at 55 days these differences disappeared. We conclude that it is possible to replace SBM in SG feed with a combination of local protein sources while maintaining performance results.

Influence of supplementing bakery by-products and cocoa bean shells to a herbage-based diet on rumen microbiota and gas emissions of early-lactating dairy cows

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The study delves into the potential of repurposing bakery products (BP) and cocoa bean shells (CBS) as sustainable feed alternatives for early-lactating dairy cows. Given concerns about rumen dysbiosis, a novel approach integrates BP with CBS in a fresh herbage-based diet. The study involved 34 early lactating Holstein and Red Holstein dairy cows across spring and autumn trials. Each trial spanned 6 weeks, comprising a baseline measurement (day before trial start), a 2-week adaptation period, and a 4-week sampling period. Three concentrate types were compared: a control concentrate (CON), a concentrate consisting of 55% BP, and a BP concentrate where straw meal was replaced by CBS (5%, BP+). Rumen fluid was collected the day before the start (T0), after 3 (T1) and 6 weeks (T2), and subsequently analyzed for microbiota characterization. Additionally, individual emissions were measured daily via a GreenFeed system. The findings revealed that feeding BP enhanced ($P < 0.05$) bacterial richness and biodiversity compared to CON, and both BP and BP+ significantly ($P < 0.05$) affected the beta-diversity in both spring and autumn trials. In the spring trial, BP led to increased ($P < 0.05$) CO₂ emissions compared to both the CON and BP+ diets, although no such differences were noted in autumn. Overall, the diet-independent, maintained integrity of the rumen microbial population underscores the viability of combining BP and CBS with fresh herbage to sustainably feed early-lactating dairy cows.

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Evaluation of ensiled fruit wastes with crop by-products as feeds for ruminants

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In regard to the circular economy, we explored the nutritional aspect of ensiled fruit wastes with crop by-products as cattle feed. First, we screened 48 ensiling treatments with 4 substrates (apple pomace, citrus pulp, mango peels and sugar beet pulp), 3 substrate locations, 2 co-substrates (straw or sugarcane bagasse) and 2 ensiling periods (1 and 3 months). Using the gas test, all substrates performed similarly when paired with sugarcane bagasse showing 63-70% estimated organic matter (OM) degradability, but apple pomace and mango peel were superior to citrus pulp and sugar beet pulp when paired with rice straw ($P < 0.05$). Next, apple pomace, citrus pulp and mango peels ensiled with sugarcane bagasse were tested against corn silage (control) in the RUSITEC system. All diets differed only in the kind of ensiled product in the composition and were similar in OM, crude protein and fat contents. The control (C) and mango peel (MP) diets contained less fiber but more non-fiber carbohydrates compared to the citrus pulp (CP) and apple pomace (AP) diets. The pH and redox potential were 6.3–6.4 and -204 – -238 mV, respectively. The C diet had 10% more OM degradation but 20% less methane formation than the fruit waste diets ($P < 0.05$). All fruit waste diets increased acetate but decreased propionate and butyrate proportions compared to C ($P < 0.05$), with MP showing the strongest effect. The MP diet decreased ammonia concentrations by 35-45% compared to other groups ($P < 0.05$). Concluding, ensiled fruit wastes could be valuable forage sources for cattle. The strong effect of mango peel on microbial metabolites indicates the influence of its bioactive compounds on rumen microbiota.

Effect of feed ingredient based on processed former foodstuff in dairy cow diet: performance and environmental impact

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Aim of the study was to evaluate the effects on performance and environmental impact of a circular diet with processed former foodstuff (FF) compared to a control diet (C). Corn meal and molasses in C were completely replaced by former foodstuff in FF. A change-over design was applied on 34 Italian Holstein cows, subdivided into two homogeneous groups. Individual milk production was recorded and individual samples were analysed for quality. Diets and feces were collected and the uNDF content was determined and used as internal marker to estimate digestibility. Environmental impact of the diets was estimated with the Simapro software. Estimated (kg/d) dry matter intake (on average 25.7) and measured milk production (39.3 kg/d) were not affected by diet. The concentrations of lactose and urea showed a significant difference between diets (4.95 in C vs 4.84% in FF for lactose; $P < 0.001$) and (26.8 in C vs 29.0 mg/dL in FF for urea; $P = 0.003$). Cows fed FF showed a significantly higher total chewing activity than C ($P = 0.020$), with higher ingestion activity (354 vs 328 min/d for FF and C) and rumination activity (575 vs 547 min/d for B and C). The estimated in vivo total tract OM digestibility was not affected by diet (76.9% on average $P = 0.557$) but there was a tendency for a higher NDF digestibility for B (47.0 vs 45.4% $P = 0.06$, respectively for FF and C). The global warming potential due to diet production (kg CO₂ eq) was 18.9 and 16.2 respectively for C and FF. In conclusion, products no longer intended for food use but safe if used as feed improved the sustainability of the milk production by reducing competition with traditional ingredients crop based.

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Environmental impact and productivity of Reggiana dairy herds fed hay or fresh forage

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The study aimed at evaluating the productivity and estimating the environmental impact of Reggiana dairy herds fed hay (H) or fresh forage (F). Nine herds (75±38 cows; 592±40 kg body weight), raised in the Parmigiano-Reggiano area (northern Italy), were monitored in summer (S) and winter (W) 2022/23 while fed F or H respectively. Environmental and managerial conditions, forages and concentrates (C) chemical composition and digestibility, productivity data and animal's intake and characteristics were acquired twice per season and entered into the nutritional dynamic software (NDS) to generate environmental impact data. Diets were 14.1±1.7 and 14.8±1.6 CP, 44.1±4.5 and 41.8±4.4 NDF and 14.6±2.7 and 15.7±3.4 Starch (% DM basis) in S and W respectively. The temperature-humidity index (THI) was different between seasons, averaging 76.9±3.5 and 54.1±7.6 respectively in S and W. Diets were on average represented by 41.3±10.9% H, 29.3±13.7 % F and 29.2±7.9% C during S (DMI=20.6 kg/d), and 65.9±5.9 % H and 34.0±5.9% C during W (DMI=17.0 kg/d). Energy corrected milk (ECM) was calculated based on milk yield and composition while N use efficiency (NUE), NH₃, CO₂ and CH₄ emissions were estimated through NDS inputted data. Statistical analysis was performed by the GLM univariate procedure, using the THI as covariate to study the forage effect. No significant differences were found between H or F based diets, with ECM of 17.8 vs 17.9 kg ($P = 0.899$), NUE of 23.5 vs 24.9 ($P = 0.684$), NH₃ of 112.3 vs 125.2 g/d ($P = 0.683$), CO₂ of 13.3 vs 12.6 kg/d ($P = 0.623$) and CH₄ of 444.5 vs 434.4 g/d ($P = 0.762$). In conclusion, the use of H or F did not affect the productivity and the estimated environmental impact of Reggiana dairy herds.

Feeding dairy ewes with a diet containing agri-food co-products: Implications on rumen metabolism

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Feeding a mixture of agri-food co-products as ingredients of a concentrate feed in replacement of soybean meal and corn grain is considered an interesting strategy in dairy ewes to improve dietary sustainability. This study aimed to assess the effect of such dietary replacement on volatile fatty acids (VFA), fatty acids (FA) concentrations, microbiota composition, and methane production from the rumen fluid of grazing dairy ewes. A total of 24 dairy ewes were allotted to CTRL (concentrate based on a mixture of barley, corn and soybean meal) or EXP (concentrate based on a mixture of olive pomace, tomato peels, distillers, wheat bran, and rice husk) groups, balanced for milk yield (1.1 ± 0.5 kg) and days in milk (82.3 ± 31.7). The trial lasted six weeks, and samples of rumen fluid were collected at the beginning and the end of the trial. Total VFA and methane productions were similar between the two groups, indicating similar fermentation patterns of the two diets despite EXP having a higher content of fibre. The presence of co-products in the EXP diet also provided a greater amount of PUFA, which determined a different biohydrogenation pattern that, in turn, resulted in higher overall concentrations of C18:1 trans-FA and a lower content of PUFA in rumen liquor. Rumen microbial community was also characterized by high-throughput sequencing of 16S rRNA gene.

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Theatre 13

Utilization of orange peels as a high value secondary feedstuff for dairy sheep

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In a worldwide context of increasing demand for animal-derived products, speculations of the feed market and climatic risks, sheep farming faces the daily challenge of ensuring its sustainability, while food by-products arise as alternative feeds, due to their nutritional value and wide availability. The aim of the present study was to evaluate the use of orange peels from a Greek orange juice industry as a high value secondary feedstuff for dairy sheep, within a circular economy approach. Enzymatically hydrolyzed (processed) and unprocessed orange peels were introduced to 2 groups (n=12/group) of dairy ewes of Chios breed at 11% DM inclusion levels (groups P & U), while group C of 12 ewes served as controls. All rations were isoenergetic and isonitrogenous and ewes were individually fed for 84 days. Milk production and composition were measured biweekly. Although fat corrected milk yield was significantly higher ($p < 0.05$) between U and C treatments on Day 14 only, at most samplings milk fat of U group was significantly higher ($p < 0.05$) than the C and milk protein was higher at all samplings for U group ($p > 0.05$). It appears that unprocessed orange peel feed could increase acetic acid production in the rumen, thus increasing milk fat content. In conclusion, orange peels could potentially be a promising secondary feedstuff that can be used as a sustainable alternative ingredient for dairy sheep nutrition. Acknowledgement: EU PRIMA Program (grant agreement n°2013).

Effect of pre and postnatal learning strategies in extinction of conditioned taste aversion for pine leaves in sheep

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Conditioned taste aversion (CTA) could prevent the consumption of leaves in sheep when are used to weed agricultural production systems. However, the extinction of the conditioned behavior may restore leaves intake if the soil plant resource is scarce. 48 lambs were submitted to a CTA protocol after weaning, where they orally received 150 mg/Kg of LiCl after the consumption of pine leaves. Previously, prenatal and social learning were applied to enhance CTA in a 2×2 factorial design; 16 lambs were prenatally conditioned (PRE), 16 lambs socially learned from conditioned ewes (SOCIAL), 16 lambs were exposed to both experiences (PRE-SOCIAL) and 16 lambs only received the CTA protocol (CONTROL). The intake of pine leaves was analyzed at extinction days 1-6 with the group, sex and their interaction as the main factors of study. The group tended to affect the pine intake only at extinction day 1 (P=0.063), where the CONTROL group tended to ate more than PRE (0.1) and PRE-SOCIAL (P=0.089) groups. Interestingly, an interaction between group*sex was observed on extinction days 4 (P=0.031), 5 (P=0.097) and 6 (P=0.016), where females ate more than males in the CONTROL and SOCIAL groups but less than males in PRE and PRE-SOCIAL groups. CTA in lams could be enhanced by prenatal and social learning observing an interaction with the sex of animals. This information could be used to improve sheep grazing systems inside agricultural production.

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Poster 15

Effects of olive cake and gender on the performances and hematology of fattening pigs

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Given the costs of animal feed and the growing demand for food, in recent years there has been a search for new sources of raw materials for animal feed. The possibility of agroindustry co-products valorization as diet ingredients, such as olive cake (OC), can reduce the environmental impact and the production costs. A trial was carried out to evaluate the effects of dietary inclusion of OC on growth performance (ADG, ADFI and FCR) and blood parameters (Gamma GT, ALT, AST, alkaline phosphatase, cholesterol and triglycerides) in commercial pigs in the final phase of fattening (127 to 148 days of age). From a basal diet, two more diets were formulated with 2.5 and 5.0% inclusion of OC, which were assigned to 24 pigs (12 males and 12 females; mean of 84 kg BW) in groups of eight, balanced for sex and weight. The results show that diet had no effect on the animals' growth performance, namely the final live weight was similar among groups (106 a 108kg). The gender had effects (P<0.05) on the performances, the males had higher final BW (110.0 vs. 103.7 kg), higher ADG (1.135 vs. 0.987 kg/day) and lower FCR (3.296 vs. 3.844). The blood parameters evaluated were not influenced by diet, but the cholesterol and triglycerides showed a tendency (P<0.1) towards a lower value in males. The results suggest that the inclusion of up to 5% of OC in diets for pigs does not negatively affect the growth performance and blood parameters of pigs in the final period of fattening. Funding: This work was also supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Effect of dietary hybrid rye inclusion on growth performance of growing-finishing pigs

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Hybrid rye is a drought-tolerant cereal variety with adaptability poor soils. It is more resistant to ergot and fusarium and has lower content of anti-nutritional factors compared to common rye. This study assessed the effect of feeding hybrid rye substituting wheat and maize grain in diets fed to growing-finishing pigs on their growth performance. A total of 528 pigs ([Landrace x Yorkshire] x Pietrain), weighing $26 \pm \text{SD } 4.6$ kg, were distributed across 48 pens based on weight and sex. The study included three phases: Growing 1 (G1; d0-21), Growing 2 (G2; d21-49), and Finishing (F; d49-105), each corresponding to different feed formulations. Two dietary groups were established: one with conventional ingredients (wheat and maize; CON), and the other completely replacing wheat and partially replacing maize with a 40% inclusion of hybrid rye (HR). Growth performance and carcass characteristics were analyzed using linear models with pen as experimental unit. Overall, growth performance was similar between groups (CON: 111.9 kg BW at d105, 0.81 kg/d average daily gain [ADG], 2.28 kg/d feed-to gain ratio [FGR]; HR: 111.4 kg BW at d105, 0.81 kg/d ADG, 2.29 kg/d FGR). In G2, pigs fed HR exhibited higher average daily feed intake (ADFI; CON: 1.82 kg/d; HR: 1.89 kg/d; $P = 0.025$) and a tendency for increased ADG (CON: 0.87 kg/d; RYE: 0.93 kg/d; $P = 0.074$). Pigs fed HR also showed lower mortality in G2 (CON: 1.17%; RYE: 0%; $P = 0.037$), resulting in a numeric difference for the entire study (CON: 3.56%; RYE: 1.52%; $P = 0.152$), possibly attributable to intestinal health effects. In addition, HR group showed higher carcass yield (CON: 79.1%; RYE: 79.8%; $P = 0.003$). In conclusion, a 40% inclusion of hybrid rye replacing wheat and maize as source of energy in pig diets may become a suitable alternative for growing-finishing periods, especially in hydric stressed regions, promoting resilient crop use.

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Poster 17

Bakery products in diets for weaned piglets

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To enhance sustainability of pig production, the concept of circularity has gained increasing attention. Circularity can be improved by replacing human-edible feed ingredients as cereal grains by alternative ingredients as by-products and food unfit for human consumption. The aim of this study was to determine the effect of total or partial replacement of wheat by discarded bakery products in the diet of weaned piglets. In total 704 piglets (weaning age 27 days) were allocated to 4 dietary treatments at D7 post-weaning. Treatments were: (1) control diet; 35% wheat; and diets with (2) 33%, (3) 67% and (4) 100% replacement of wheat with bakery products. These diets were fed from D7-D28 post-weaning. Data were analysed with ANOVA using Genstat statistical software with pen as the experimental unit. Replacing wheat with bakery products tended ($P=0.06$) to linearly reduce feed intake from 595 to 561 g/d, without significant effects on daily gain and feed efficiency. Total replacement by bakery products resulted in an increased incidence of diarrhoea at D9 and D12 post-weaning ($P<0.01$). Replacing wheat by bakery products linearly increased the digestibility of dry matter, organic matter and NSP and linearly decreased digestibility of crude fat and ash. Replacing wheat by bakery products reduced the carbon footprint of the diets by 32%, i.e. from 987 to 673 g CO₂ equivalents including land-use change (LUC) per kg of feed. Diet composition did not affect efficiency of N-and P-utilisation. These results indicate that wheat can be completely replaced by bakery products with minor effects on growth performance and without compromising animal health, while faecal consistency may be transiently reduced.

Seaweeds: An Alternative Feed Source in Bovine Production in the Azores

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The Azorean dairy production system is characterized by pasture-based feeding, due to the excellent edaphoclimatic conditions in the region, accounting for over 30% of Portugal's milk production. However, there are periods when pasture productivity decreases, necessitating supplementation with other feed. Considering the substantial biomass of macroalgae in the Azorean Sea, these could be a sustainable and innovative feed supplement for Azorean cattle during periods of reduced pasture productivity. This study aims to investigate the nutritional potential of four marine macroalgae, *Ulva rigida*, *Ulva compressa*, *Asparagopsis taxiformis*, and *Asparagopsis armata* as feed supplements in the Azorean livestock production system. Three samples of each of these macroalgae were collected on Terceira Island, washed in freshwater, and dried at 65°C. The samples were analyzed in triplicate for dry matter (DM), crude protein, ether extract, and total ash, according to AOAC standard methods (1995). Fibrous fractions (NDF, ADF, and ADL) were determined according to Goering and Van Soest (1970). All macroalgae had DM values below 15%, with a minimum value of 6.55% found in *A. taxiformis*. The protein content of the four macroalgae species studied ranged between 22.69 and 25.76 g/100g DM. Variations in cell wall constituents were also observed among the four algae, with *A. taxiformis* showing a high hemicellulose content, as well as values exceeding 10g/100g DM of ADL in *A. armata* and *U. compressa*. This study demonstrates that the studied macroalgae have the potential to be integrated into the diet of Azorean cattle, specifically as a protein supplement during periods of pasture scarcity when animals are supplemented with roughage feed.

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Poster 19

Nutritional and functional evaluation of *Euglena gracilis*, *Porphyridium purpureum* and *Arthrospira platensis* for feed applicationE. Petrosillo¹, D. Lanzoni¹, R. Rebucci¹, V. Perricone¹, A. Agazzi¹, G. Savoini¹, F. Cheli¹¹ University of Milan, Department of Veterinary Medicine and Animal Sciences, Via dell'Università 6, 26900 Lodi, Italy

Protein-energy malnutrition has prompted scientific research to study new protein sources for the feed sector that can guarantee a high nutritional profile and environmental protection. *Euglena gracilis* (EG), *Porphyridium purpureum* (PP) and *Arthrospira platensis* (AP) are microalgae of great interest in feed industry due to wide range of potential applications. The nutritional aspect of EG, PP and AP are well-defined literature but their functional aspects require further investigation. This study aims to evaluate the total phenolic content (TPC) by Folin-Ciocalteu and antioxidant activity (AA) by ABTS and FRAP methods of EG, PP and AP, following chemical extraction (H₂O:EtOH, 50:50). Results showed that EG exhibited a significantly (p-value<0.05) higher TPC (988.23±24.40mg Tannic Acid Equivalent (TAE)/100g) than AP (730.12±104.12mg TAE/100g) and PP (352.5±63.74mg TAE/100g). Same trend was also confirmed for ABTS (3664.08±301.73mg Trolox Equivalent (TE)/100g; 2358.29±183.8mg TE/100g; 1318.79±127.99mg TE/100g, for EG, AP and PP, respectively). FRAP was higher for both AP (58.10±5.32mg FeSO₄) and EG (49.24±1.35mg FeSO₄) than PP (33.06±4.60mg FeSO₄). Results showed that EG has a higher AA than AP and PP but further investigations on other functional profiles, specifically after in vitro digestion are necessary to determine the optimal microalgae species for application in feed sector. This study was carried out within Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022).

Evaluation of the in vitro Antioxidant Capacity of Microbial, Plant-based and Insect Protein Sources

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Due to population growth and climate concerns, traditional protein sources are insufficient. Research has identified microbial proteins as sustainable alternatives with high nutritional quality and lower environmental impact. The aim of this study was to evaluate the antioxidant activity of different protein sources: traditional (TR) (beef, whey, egg, cod); plant-based (PB) (soy, lupine, hemp, pea); insect (Acheta domesticus (Ad), Hermetia illucens (Hi)); microbial (MB) (Spirulina (Sp), Chlorella (Ch), Saccharomyces cerevisiae (Sc)). Antioxidant activity was measured with ABTS, FRAP and ORAC methods, following a biphasic chemical extraction (H₂O:EtOH, 50:50) and an in vitro digestion (INFOGEST). Data (mean±SEM) were analyzed with one-way Anova using GraphPad Prism 9 (p-value<0.05). For FRAP and ABTS the results showed that MB proteins, compared to TR and PB proteins, generally had a significantly greater values (Sp (62.27±2.20mg FeSO₄), Sc (2047.83±145.64mg Trolox Equivalent (TE)/100g)). On the other hand, Ad (233.24±6.01mg FeSO₄/100g; 3839.05±245.99mg TE/100g) showed a statistically significant values compared to all protein categories. ORAC results highlighted that MB, compared to TR and PB proteins, had a different trend compared to ABTS and FRAP methods. In particular, Ad (175.41±43.15mg TE/100g), lupine (199.01±4.48mg TE/100g), Hi (195.34±19.11mg TE/100g) and beef (242.77±0.65mg TE/100g), showed significant values compared to all other proteins tested. While the data indicated that MB showed relevant antioxidant value than TR and PB, more research will be required to demonstrate the benefits for use in the food and feed sector.

Chemical and in vitro pineapple by-products characterization for sustainable small ruminant production

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Pineapple fruits processing generates different wastes which are thrown away and contributes to environmental pollution. In this study pineapples waste for small ruminant nutrition have been characterized. Five by-products (crown, bud end, peel, core, and pomace) from two pineapple varieties (Smooth Cayenne_SC and Sugarloaf_SL) were collected to determine the chemical composition, estimate the metabolizable energy, and study the in vitro fermentation characteristics using sheep as donor animals of rumen fluid. Data were statistically analysed. The results revealed a low dry matter (DM) content (on average SC 17.3 % ± 2.37 and SL 18.1 % ± 2.84). The pomace showed the highest (p<0.001) protein content (8.10 and 8.81 % DM, for SC and SL, respectively), whereas crown had the highest (p<0.001) fiber amount (NDF: 47.62 and 39.01 % DM, for SC and SL, respectively). Core and pomace showed the highest (p<0.001) metabolizable energy contents (ME, MJ/kg: 7.8 and 7.9 in core, 8.0 and 7.4 in pomace), the highest (p<0.01) organic matter degradability (OMD, %: 84.8 and 86.5 in core, 85.4 and 81.4 in pomace), and total volatile fatty acid (VFA, mmol/g: 103.4 and 109.5 in core, 90.3 and 71.7 in pomace) for SC and SL, respectively. Our finding suggests that pineapple by-products could be used in small ruminant nutrition considering crown, bud end, and peel as fiber source and core and pomace as concentrate feedstuff.

Herbal supplementation in dairy cows' diet: impact on rumen, milk and cheese microbiota

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The study aimed to evaluate the effects of including *Echinacea purpurea* (EP) and *Galium odoratum* (GO) in cows' diet, on rumen microbiota and on microbiological quality of milk and cheese. EP was chosen in a circular economy view using the plant residues obtained from the production of the mother dye, in this process aerial part is considered a waste and a cost for industries. Several studies showed an association between EP supplementation, immunomodulation and reduction of inflammation. GO was chosen for its diuretic, anti-inflammatory properties and for its prevalence in Vezzena Plateau's pastures, an area that could be restored with GO after the Vaia Storm damage. A group of 9 dairy cows were fed with a total mixed ration (TMR) diet for 3 weeks. In the second week 3 out of the 9 cows were supplemented with EP and 3 with GO. Herbs were gradually added to the daily TMR to get the dairy cows used to the new feed avoiding any negative effects on feed intake, rumen environment or milk production. Individual milk and rumen liquid samples were collected from each cow. Milk samples were used for experimental production of mini-cheeses (about 200g each one). Milk and cheese were homogenized, decimally diluted and plated for the research of coliforms, mesophilic and thermophilic lactic acid bacteria. Microbial DNA from rumen samples was extracted and sequenced by MiSeq Illumina NGS methodology. As preliminary results, the supplementation of herbs didn't have a significant impact on milk or cheese microbial population. An increase in *Verrucomicrobia* and *Fibrobacter* taxa was observed in rumen microbiota after GO supplementation. *Verrucomicrobia* taxa are about 1% of rumen microbiota, they are found in diets rich in "low quality" forage. *Fibrobacter* taxa are important fiber degraders and volatile fatty acid (VFA) producers. Results obtained on ruminal microbiota suggest a potential use of these plants in cows' diet.

Chemical characterization and phenolic content of winery and grape by-products as potential feed supplement

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Circular economy has received significant attention for considering both agro-industrial sustainability and socio-economic concerns, seeking for innovative solutions for waste disposal. The agri-food sector stands out for its production of by-products, many of which are used as animal feed. Grapes are one of the most cultivated fruit crops globally, with approximately 67 million tons produced annually. These sectors generate up to 20% waste biomass in the form of grape skin, seeds, stems, and residual pulp, known as grape pomace. The potential benefits of grape by-products (GP) provided by bioactive compounds deserve exploration when considering their addition in formulations. Moreover, they seem to potentially reduce enteric methane when fed to ruminants. The aim of this study was to analyse seven grape by-products (GP1 to GP7) to determine their chemical composition, polyphenolic content, as well as flavonoid and anthocyanin profiles. All GP showed an excellent content of PG (18-23%) and EE content, mostly in GP2 (12.11 %) as seed part. Fibre content aligns with literature, with NDF ranging from 25.52 to 48.80 and ADF from 27.80 to 32.19 (DM basis). The analysis reveal that GPs possess rich polyphenolic content (2.90-17.72 mg/g GAE) sequenced as GP7>GP6>GP5>GP4>GP3>GP2>GP1 and anthocyanin (278-2810 mg/kg) sequenced as GP1 >GP3>GP5>GP4>GP7>GP6>GP2. These characteristics alongside nutritional value suggest their potential as valuable supplements in animal feed aligning with the principles of circular economy and offering both environmental and economic benefits.

Effects of the inclusion of grape stem at different rates in intensive fattening cattle diets on in vitro fermentation parameters

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The inclusion of by-products that do not compete with human food in ruminant diets is encouraged. Grape stems may be of interest because they are rich in polyphenols and proanthocyanidins, despite their low crude protein (CP) and high content of fibre. The aim of the present study was to evaluate the effect of replacing straw with different proportions of grape stems (0%, 3%, 6% and 9%) in two beef cattle concentrates differing in CP content (12 vs 14%) on in vitro fermentation parameters: gas, methane (CH₄), dry matter degradability (IVDMD), ammonia (NH₃-N), and volatile fatty acids (VFA). For that, bovine ruminal fluid was used to evaluate 8 mixtures in an in vitro fermentation assay with Ankom system (3 replicates in 3 separate runs) during 48 h. The diet consisted of 91% concentrate (12% or 14% CP) plus 9% of stem/straw combination (0/9, 3/6, 6/3, 9/0 % of stem/straw). The inclusion of grape stem in the diets with 12% CP concentrate caused a linear increase in gas (P=0.01) and ammonia production (P<0.05) without affecting CH₄ production, IVDMD, and total VFA (P>0.05). The inclusion of grape stem had a quadratic effect on the proportion of acetic acid (P=0.009), butyric acid (P=0.04) and valeric acid (P=0.008), while it linearly decreased the proportion of propionic acid (P<0.001). On the other hand, in the diets with 14% CP concentrate, the inclusion of grape stem only linearly increased the proportion of acetic acid (P=0.001) and linearly decreased the proportion of propionic acid (P<0.001). These results support the substitution of straw by grape stem in fattening diets, especially when high-protein concentrates are used.

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Poster 25

Avocado (*Persea Americana*) seed as an alternative ingredient in the diet of replacement goats. In vivo digestibility, N balance and energy balance

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Avocado industry is rapidly expanding, yielding over 100,000 tons/year of fruit in Spain and generating substantial wastes. These needs exploring sustainable options like using these by-products in livestock feed. The avocado seed (AS) shows promising properties in this way. Therefore, this study aims to assess the impact of incorporating AS into the diets of young goats, focusing on feed intake, digestibility, energy/N balance and metabolic indicators. Twenty Murciano-Granadina goats aged six months (BW = 25.5 ± 1.7 kg) were divided into two groups, the control (CTL) group (n=10), fed with a commercial concentrate and the treatment (AVC) group (n=10) fed with the same concentrate where a 9% of barley and 2% of maize were replaced by AS flour. Both groups were offered 80:20 forage to concentrate. After a 15d adaptation period, animals were placed individually in metabolic cages (5 d) for nutrients digestibility assay. Daily intake was higher in CTL than AVC (1.06 vs 0.96 kg, P=0.027). No significant differences resulted in DM, OM, CP, CF, and ADF digestibility (P>0.05) between CTL and AVC. However, CTL showed lower NDF intake (295 vs 349 g/d, P=0.002) and digestibility (37.0 vs 45.9 %, P=0.004) than AVC. N excretion was higher in CTL (1.28 vs 1.11 g/kg BW^{0.75}, P=0.051). The CTL had higher digestible energy relative to energy intake than AVC (75.7 vs 65.6 %, P=0.030). No significant differences were obtained in plasmatic β-Hydroxybutyrate concentration (P>0.05). Yet, systemic NEFA concentration was higher in CTL than AVC (0.332 vs 0.268 mmol/L, P=0.045), possibly due to higher crude fat intake in CTL (31.2 vs 24.8 g/d, P<0.001). No significant differences in urine purine derivatives (P>0.211) indicated unaffected microbial protein synthesis. Adding AS to young goats' diet had no adverse effects on N use, nutrients digestibility, metabolism, or rumen microbial protein synthesis. A balanced energy and ruminal nitrogen supply are crucial for successful AS inclusion in ruminant diets, making it a suitable ingredient for circular economy and livestock sustainability.

The impact of incorporating avocado (*Persea Americana*) seed into the diet of replacement goats on ruminal fermentation and blood parameters

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The growing avocado use in Spain generates significant by-products, including avocado seed (AS). Utilizing AS in the agri-food industry supports circular economy and livestock sustainability. Our unpublished findings suggest promising properties for goat feeding in terms of nutrient digestibility, N and energy balance, and microbial protein synthesis. This study assesses AS inclusion effects on ruminal fermentation (VFAs, pH, ammonium) and blood parameters in young Murciano-Granadina goats. Twenty goats (BW = 25.5 ± 1.70 kg) were split into control (CTL) and treatment (AVC) groups, with AVC receiving AS flour replacing 9% barley and 2% maize. After a 15-day adaptation and 5-day digestibility assay, blood and ruminal fluid samples were collected for analysis. No significant differences were found in ruminal pH, ammonia-N and total VFA concentration between CTL and AVC (P>0.05). However, the Ac/Pr (2.77 vs 1.87, P=0.03) and acetic molar proportion (58.7 vs 51.8, P<0.001) was higher in the AVC, which may be caused by the lower intake of this group compared with CTL (962 vs 1060 g/day, P=0.027). The haematological parameters showed that the cholesterol level in blood was significantly higher in the CTL than in the AVC (60.6 vs 28.9 mg/dL, P<0.001). However, the level of GPT was higher in the AVC compared with the CTL (31.2 vs 19.1 U/L, P<0.001), which could indicate a possible occurrence of hepatic toxicity in the event that the inclusion of AS was increased above the percentage studied. Finally, the levels of urea were lower in the CTL than in the AVC (29.3 vs 35.4 mg/dl, P=0.004) which may be caused by the recirculation of N destined to become protein, which could also be observed numerically in the lower N excretion in feces and urine by the animal. No other significant differences were observed in the blood profile. Using AS in young goats' diet had no negative impact on ruminal fermentation. Increased GPT and blood urea levels may suggest potential liver overload and protein metabolism redirection. Exploring lower AS percentages (<12%) is relevant for assessing its impact on animal feed.

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Poster 27

Can the use of dietary oilseeds improve the nutritional and sensory properties of Podolian x Limousine meat?

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The decrease in SFA and the increase in health beneficial PUFA is a main topic of meat and meat products research. The present study investigated the effects of different oilseeds supplementations on nutritional and sensory quality of bovine meat. Thirty two Podolian x Limousine young bulls were randomly allocated into four experimental treatments: control diet (CO) with no supplemental fat, linseed diet (LS) with a supplementation of 10% whole linseed; sunflower diet (SS) with supplementation of 10% of sunflower seeds; LS+SS diet: with supplementation of linseeds (5%) and sunflowers (5%). After finishing period, all animals were slaughtered, fatty acids profile, nutritional indices and sensory proprieties were evaluated on meat. All data were subjected to an analysis of variance using the GLM procedure of the SAS statistical software. Results highlighted that supplementation with LS, SS And LS+ SS decreased the percentage of saturated fatty acids and increased monounsaturated and polyunsaturated fatty acids. Feeding diets containing LS versus SS increased the amount of total PUFA n-3 and of individual PUFAs n-3 as linolenic, eicosapentaenoic, docosapentaenoic and docosaeasenoic. An improvement in tenderness, juiciness, chewiness and flavour intensity was observed in beef from animals supplemented with oilseed. Data from the present experiment highlighted that linseed alone or in combination with sunflower seeds is an effective strategy to improve fatty acids profile and some sensory attributes in beef.

Effects of dietary supplementation with enriched olive cake on growth, rumen fermentation, and metabolic status in Limousine bulls

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Inclusion of olive cake (OC) into animal diet is an opportunity for circular economy, animals' health, and animal products quality. The aim of this study was to evaluate the effects of dietary polyphenols enriched OC (EOC) supplementation on performance, inflammometabolic response, and rumen metabolism of Limousine bulls. Thirty bulls were blocked by body weight (BW) and then randomly assigned to 1 of 2 dietary treatments for 80 d: concentrate without inclusion of EOC (CTR) and concentrate with 13% inclusion of EOC (EOC). BW measures and blood samples were collected at 0, 40 and 80 d of treatment, whereas rumen fluid samples were collected after slaughtering. Data were analyzed with the GLIMMIX procedure of SAS. EOC bulls had greater rumen propionate and lower acetate, isobutyrate, and isovalerate content, as well as lower acetate:propionate and acetate+butyrate:propionate ratio than CTR bulls. EOC group showed greater levels of blood urea, γ -glutamyl transferase, and alkaline phosphatase, and lower levels of fructosamine, albumin, ferric reducing antioxidant power, calcium, and zinc than CTR group. Although no differences between groups were achieved for performance, the inclusion of 13% of EOC in the concentrate could result in a worsen feed efficiency (high content of low-degradable NDF and ADF) and lower liver functionality due probably to high level of organic peroxides of the enriched OC supplement. However, the lower rumen acetate and the greater propionate concentration in the EOC bulls could be indicative of a positive modulation of rumen metabolism towards lower methane production.

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Poster 29

In vitro rumen fermentation of forage-based diets supplemented with hydroxytyrosol extracts

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Dietary polyphenols may modify rumen fermentation pathways. In vitro gas production and fatty acids (FA) were analyzed after adding an hydroxytyrosol (HT) extract (0 vs. 178±36 mg/kg) to forage-based total mixed rations (TMR) (50% neutral-detergent fiber; 11.4% crude protein; 2.1% ether extract with 37.1% of C18:2 n-6 and 9.7% of C18:3 n-3). Rumen fluid was collected from twelve slaughter cows and each sample (n=8 per treatment) was incubated in triplicate in three separate runs. The dried matter (DM) samples were incubated with rumen fluid (pH set at 6.9±0.11) in a water bath at 39 °C for 48 h. Data were analyzed with mixed models including diet as fixed effect and incubation run as random effect. The final pH did not differ between diets (6.64 vs. 6.67±0.035, in control and HT, respectively; p>0.10). Gas production after 24 h incubation tended to increase by HT addition (79.8 vs. 84.1±5.54 ml/g incubated DM; p=0.10), but it did not differ after 48 h (99.2 vs. 100.7±4.41 ml/g incubated DM; p=0.76). Both in vitro DM disappearance and true DM digestibility were enhanced by HT addition (25.7 vs. 33.2±2.15% and 34.7 vs. 42.5±1.93%, respectively; p<0.01). Total FA content in residues did not differ across treatments (70.3 vs. 72.5±7.57 mg/g DM; p>0.10). Biohydrogenation of C18:2 n-6 and C18:3 n-3 was similar across treatments (96.4 vs. 94.7±2.27% and 98.3 vs. 97.1±0.98%, respectively; p>0.10). The only differences in lipid composition were observed in the sum of dimethylacetals, which were higher by adding HT (0.29 vs. 0.31±0.042%; p<0.05). Adding HT to TMR increased rumen degradability of structural carbohydrates but had minor effects on unsaturated FA hydrogenation. Funded by PID2020-113617RR-C21.

Evaluation of formic acid as additive for almond hulls silage

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There is limited information on the ensiling potential of almond hulls (AH), the main byproduct of almond production. Formic acid (FA) is frequently used as silage additive, as it can improve silage quality for a wide range of crops. This work aimed to analyze the effects of FA on quality and microbiology of AH silage. Fresh AH (60,7% dry matter (DM); on DM basis: 7,55% crude protein; 38,7% neutral detergent fibre) was hand-cut (about 1 x 1 cm), and 500 g of AH were weighed into each of 10 vacuum bags. In 5 bags FA was added (2 ml of FA/bag diluted in 107 ml water) and the other 5 bags (Control) received only the same volume of water. Bags were vacuum packaged and stored in darkness at room temperature. After 3 months of storage, bags were opened and their content analyzed for pH, DM and lactic acid content. Microbiology of silage was assessed by classical plate-culture techniques. Compared with control silage, the addition of FA reduced pH of silage from 5.04 to 4.56 ($P < 0.001$) and decreased DM losses from 2.31 to 1.46% ($P < 0.001$). However, FA decreased lactic acid concentrations from 10.5 to 1.24 g/kg DM silage ($P < 0.001$). No *Escherichia coli*, *Salmonella* spp., *Listeria* spp., *Clostridium* perfringens and *Staphylococcus* coagulase-positive were detected in any silage. Whereas no presence of yeast was observed by adding FA at ensiling, yeast were present in all control silages (4.66 log CFU/g silage as average). Using FA also reduced mold counts from 2.80 to 1.31 log CFU/g; $P = 0.002$), but had no effect on *Enterobacteria* spp. (1.20 and 0.95 log CFU/g; $P = 0.193$). The results indicate that using FA as silage additive can improve the microbiological quality of AH silage and reduce DM losses.

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Poster 31

Variability in energy content and protein value for ruminants of brewers' grains from different breweries

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Brewers' grains (BG) are the most abundant by-product of the brewing industry and are widely used in ruminant feeding. This work aimed to estimate the energy content and protein value of 14 samples of BG from different Spanish breweries. Samples were dried (40°C), analyzed for chemical composition and incubated in vitro with buffered ruminal fluid from 4 sheep for 24 h to measure gas production. In addition, 3 Ankom bags with each BG sample were incubated with buffered sheep ruminal fluid for 16.5 h (6% rumen passage rate) to determine dry matter (DM) and crude protein (CP) degradability. Metabolizable energy (ME) content was estimated from chemical composition and gas production at 24 h. Protein value (PDIA, PDIN and PDIE) was calculated using INRA 2007 equations and in vitro CP degradability. Content of CP ranged from 24.2 to 32.8 g/100 g DM (average 28.1%), and CP degradability varied from 25.4 to 66.5% (average 49.3%), being these values in good accordance to those reported by Feedipedia but greater than that of INRA (45.0%). Average values of PDIA, PDIN and PDIE (13.2, 26.5 and 19.9% DM, respectively) were greater than those of INRA (13.7, 19.4 and 17.7% DM), which agrees with the greater CP content in Spanish compared with French BG (28.1 vs. 26.2% DM, respectively). Average ME content (8.9 MJ/kg DM; 8.15 – 9.58 MJ/kg DM) was similar to the 9.0 MJ/kg DM value of Feedipedia estimated from chemical composition and gas production. However, values were lower than that of INRA (9.9 MJ/kg DM), which is in accordance to the reported underestimation of ME content of high-CP feeds when using gas production as predictor. The results indicate high variability in both energy and protein value of BG.

Effects of drying and pelleting on ruminal fermentation of brewers grains

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Brewer's grains (BG) are widely used in ruminant feeding, but its low dry matter (DM) content hinders its storage and preservation. This study evaluated the effect of low-temperature drying using solar panels and subsequent pelleting (DPBG) on chemical composition and *in vitro* ruminal fermentation of BG. A total of 6 samples (3 fresh BG and 3 DPBG) were analyzed for chemical composition and incubated *in vitro* with buffered ruminal fluid from 4 sheep for 24 h to measure fermentation parameters (pH, volatile fatty acids (VFA) and NH₃-N). In addition, 3 Ankom bags with each sample were incubated with buffered sheep ruminal fluid for 16.5 h (6% rumen passage rate) to determine DM and crude protein (CP) degradability. There were no effects of treatment on either chemical composition ($P \geq 0.172$) or DM degradability ($P = 0.648$; 34.2 vs. 36.1% for BG and DPBG, respectively), indicating no negative effect of drying and pelleting. However, DPBG reduced ($P = 0.028$) CP degradability of fresh BG from 53.0 to 42.7%, likely due to the high temperature during the pelleting process that can cause protein denaturation and thus reducing CP degradability. Drying and pelleting of BG increased ($P = 0.011$) total VFA production by 21.9%, resulting in greater ($P = 0.033$) propionate proportion than for fresh BG, with no differences ($P \geq 0.106$) in molar proportions of the other VFA. As a consequence, acetate/propionate ratio was lower ($P = 0.018$) for DPBG compared with fresh BG (1.75 and 2.03 mol/mol, respectively). No differences ($P = 0.465$) were detected in NH₃-N concentrations after 24 h of incubation. Overall, the results indicate that drying and pelleting can increase BG ruminal degradation, but CP degradability can be reduced.

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Poster 33

Variability in chemical composition of wet brewers' grains from a single brewery plant

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The brewing industry produce large quantities of by-products than can be used in ruminant feeding, but there is limited information on their variability. This work aimed to analyze the variability in chemical composition of 18 samples of wet brewers' grains (WBG) obtained from a single Spanish brewery plant in different periods and from different batches. Samples were dried at 40°C for 48 h, ground to pass a 1 mm screen, and analyzed for chemical composition to determine dry matter (DM), ash, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), ether extract (EE) and water-soluble carbohydrates (WSC). The DM content ranged from 11.0 to 17.3% (average 14.5%), whereas ash ranged from 3.01 to 4.57% and CP from 21.5 to 33.5%, with average values of 3.56 and 26.7% respectively (all values on DM basis). The average content of NDF, ADF, EE and WSC was 56.8 (49.1 – 64.9%), 21.0 (18.7 – 25.1%), 10.1 (8.31 – 11.24%) and 5.95% (1.83 – 14.38%), respectively. Coefficient of variation (CV) ranged from 8 to 10% for all fractions, excepting for WSC (56.1%) and DM (12.9%). The DM content of the samples were lower than those reported by Feedipedia (21.8 – 28.9%; average 24.9%). Both WSC and EE contents were higher compared to the data reported in INRA-CIRAD-AFZ Tables (average values 1.3 and 8.6%, respectively) and in Feedipedia (1.0 and 7.0%, respectively). In contrast, ash, CP, NDF and ADF content was similar to those in INRA-CIRAD-AFZ Tables (4.5, 26.2, 57.3 and 22.1%). The results indicate high variability in chemical composition of WBG from the same brewery plant and differences in DM, WSC and EE content in comparison to the available data. Variability can be due to differences in the brewing process to produce different beers.

Effect of mineral and organic fertilisation on the biodiversity and nutritional value of Azorean pastures

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A comparison between conventional inorganic and organic fertiliser produced from residues resulting from carcass cutting on pasture biomass and its nutritional value. 25 plots (4m²) from a biodiverse pasture were ploughed, seeded with perennial ryegrass (*L. perenne* L) allocated to 5 treatments-control, inorganic and organic fertiliser (100 and 200 kg N ha/year), in a Latin Square experimental design. Effects of treatments on biodiversity, digestibility and frequency of species were tested by one-way ANOVA followed by Tukey test. During a year, pasture samples were taken monthly to access in vitro digestibility and the biodiversity was evaluated in the field once a season. In addition to ryegrass other species emerged in pasture (white clover –*T. repens* L, velvet grass – *H. lanatus* L). Treatments only affected biodiversity in summer by decreasing in control comparing to inorganic 100 and organic 200 (p = 0.08). Likewise, the treatments only slightly decreased the digestibility in the winter in inorganic 100 compared with inorganic and organic 200 (p=0.07). Concerning the frequency of species, this only was affected by treatments in summer (increased white clover in control and organic 100; p<0.001; perennial ryegrass greatly decreased in control and increased in inorganic 200; p<0.001) and in autumn (white clover increased in control and decreased in inorganic 100 and 200; p=0.002; perennial ryegrass decreased in control p=0.023). Digestibility also increased with the biodiversity (Shannon index), especially in spring (r = 0.486; p= 0.014). In conclusion organic fertilizer can increase biodiversity which by turn can increase pasture digestibility.

Session 90

Poster 35

Nutrients recycling to soil by grazing beef cattle in Catalonia region

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The objective of the study was to determine the N and mineral recycling of grazing beef cattle in the Northeast of Spain to adjust pasture fertilisation. Twelve Limousine cows (650 ± 47.8 kg of BW) were allowed to graze in 4,800 m² of mix pasture (*Bromus* spp., *Festuca arundinacea*, *Trifolium repens*, 12.8% CP and 58.3% NDF) for 12 days. Every day all animals were moved to a new 400 m² parcel, and they were twice daily dosed with a C32 alkan bullet. After 6 days of external C32 dosing, faecal and urine samples were collected to estimate feed intake at the same time as bullet dosing. Faecal output was estimated using acid insoluble ashes as internal marker. Pasture and feed samples were analysed for N, Ca, P, Mg, K, and alkans content, and urine samples for N, Ca, P, Mg, and K content. The estimated intake was 6.1 and 8.4 kg of DM pasture for pregnant and lactating cows, respectively, and both were supplemented with 1 kg concentrate feed (14% CP) during the bullet dosing. N intake was 162 and 212 g/d, N faecal excretion was 35 and 47 g/d, and N urine excretion was 65 and 73 g/d, in pregnant and lactating cows, respectively. Therefore, 61 and 56 % of the total N intake, of pregnant and lactating beef cattle returned to soil or volatilized as ammonia form. The excretion of Ca, P, Mg, and K in manure (faeces and urine) was 31 and 43, 17 and 23, 12 and 14, and 158 and 201 g/d in pregnant and lactating cows, respectively. This represented 62, 61, 76, and 84 % of Ca, P, Mg, and K intake, respectively. This information can be useful to adjust pasture fertilisation, specially, when grazing management includes small parcels with high stock density.

Effect of diet and season on Reggiana dairy cattle milk production and quality

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This study aimed at evaluating the effects of diet and season on milk yield (MY) and composition of Reggiana dairy cows. Nine dairy farms in the Parmigiano-Reggiano area were included in the trial (79±4 cows; 593±4 kg BW). Animals were fed hay and concentrate separately, and fresh forages were provided in summer (S). Sampling visits, focused on lactating cows, were conducted over 2 consecutive d, twice per season during S and winter (W). Temperature Humidity index (THI) was recorded, forages and feedstuff in the 1st day, and their refusals in the 2nd sampling day, were sampled, weighed, and chemically analysed. Dry matter intake (DMI) and MY were measured. Two bulk milk samples were collected every 12 h (at 6 am and 6 pm) on the 2nd day. Milk quality and rheological properties were determined by Milkoscan™. Animals' characteristics, environmental and managerial conditions, feeds chemical composition and digestibility were inputted into the Nutritional Dynamic System software, to evaluate dietary characteristics. The diets were on average 14.4±1.6% CP, 42.1±6.1% aNDFom and 14.8±3.3% starch (DM basis). Data were statistically analyzed by univariate procedure of the GLM. The THI differed in the 2 seasons, being 76.9 in SU and 54.2 in WI. No seasonal difference was found on DMI, MY and dietary content, except for higher peNDF and sugars, and lower crude fiber and starch in S. Milk fat, protein, lactose, and total solids were lower in the S season (P<0.05), and milk coagulated faster with better curd firmness (P≤0.001). However, when the THI was used as a covariate in the data analysis, only rennet coagulation time differed by season (13.35 vs 19.61 in S and W respectively; P=0.003).

Further exploration of calcium oxide to improve the quality of indigestible feeds

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Canada produces approximately 82.4 million tonnes of crop residues each year, representing a significant source of unused feed for livestock. Alkali treatment of these crop residues has been shown to improve their rumen degradability; however the application of these alkali treatments like calcium oxide can pose significant economic and labour costs. The objective of this study was to examine the effect of calcium oxide powder inclusion in either a barley straw or corn silage based diet during the backgrounding and finishing phase in feedlot steers. Feed intake, growth performance, feed efficiency, nutrient digestibility, stress indicators, and economics were assessed. Dry matter intake was not affected (P > 0.34) by forage source or treatment. Calcium oxide treatment decreased (P = 0.01) feed efficiency in the backgrounding but not finishing diet. Hot carcass weight was decreased (P < 0.01) by barley straw and calcium oxide treatment. In conclusion, calcium oxide administered in powder form to beef cattle consuming both high forage and high grain diets did not improve feedlot performance, carcass characteristics or nutrient digestibility.

Session 91

Theatre 1

Young EAAP session: project writing and speed dating

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Young EAAP session part I: project writing From the level of master's thesis projects to post-doctoral research, being able to write a successful project is crucial. Proper and structured proposal writing helps to develop research ideas and is essential for acquiring funding. Few educational programs include teaching project writing skills. In this year's Young EAAP session, we offer an afternoon full of interesting insights and sharing of experiences on project writing from MSc to postdoctoral / assistant professor level. In the first part of the session, we will get to know one another in an informal setting. In the second interactive part, we will initiate discussion to share experiences on project writing by organizing discussion groups around several topics. The third part of the session consists of invited researchers that will touch upon topics such as how to conceive a good idea, how to find funding opportunities, how you can move proposal from "good" to "excellent". The talks will also include the evaluators' point of view on what they search for in an application and how to meet evaluation criteria. The last part consists of a Q&A with invited speakers answering the final questions. All levels of "Young" researchers are welcome, don't hesitate to participate! Young EAAP session part II: speed dating During the speed dating event, young researchers will have the opportunity to meet experienced academics and industry representatives. You will be able to network and ask questions in small groups. Further communication of whom and where you can meet will be given close to EAAP2024 via e-mail and social media.

Session 92

Theatre 1

Guidelines for designing pig trials on performance, digestibility, meat and carcass quality

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The quality of data derived from experimental research depends upon how the experiment is designed (including factors like the number of replicates, blocking, and randomization) and how the experimental procedures are performed (such as sampling techniques). Within the PIGWEB project, one aim is to enhance the experimental design of pig trials, particularly for performance, digestibility, meat and carcass quality trials. Therefore, guidelines will be developed to offer a robust experimental framework, thereby promoting the quality and compatibility of pig research data. As a first step, an inventory of existing guidelines related to experimental design in (pig) research in web applications, journals, and books was created. This was done through a desktop study and by questioning pig research experts. Key resources and most common recommendations on e.g. sample size calculation, randomization, or reporting the experimental set-up will be presented and consolidated. This work is part of the PIGWEB project, which has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

Common mistakes in experimental design and analysis of pig trials: an overview

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An overview of the most common mistakes in experimental design and the statistical analysis of the data will be given, both in experimental design and data analysis. The main focus in experimental design will be on pig research, but the principles are also useful for research in other (farm) animals. Topics that will be discussed include randomization, blocking, experimental unit versus observational unit, replicates versus repeated measures. Discussion on the most common errors in statistical analysis and the interpretation of the results include translation of the research question into the correct hypothesis and statistical analysis, power, random effects, significant versus relevant, effect of outliers and interactions. This work is part of the PIGWEB project, which has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

Identifying key pitfalls in experimental design of pig trials: a group discussion

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Within the PIGWEB project, pig research experts were questioned about the experimental design of pig trials focusing on performance, digestibility, carcass and meat quality. Various topics were explored in an online survey, including significance level, statistical power, blinding, exclusion and inclusion criteria, experimental unit, blocking, and randomization. Additionally, common pitfalls in experimental design of pig trials were questioned. In this group discussion, our goal is to consolidate the survey results and define crucial aspects within experimental design of pig trials. This work is part of the PIGWEB project, which has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

Improving research quality and collaboration by developing standard operating procedures

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European Pig Research Infrastructures (PRI) are increasingly collaborating, reinforcing the need for robust and reproducible data collection to ensure high quality research. In addition to standardisation of analytical methods and reporting of appropriate validations, standardisation of Specific Experimental Procedures (SEP) is equally important as they can affect the quality of results. For instance, different sampling methods may involve different degrees of invasiveness and have differing impacts on behaviour and emotional states, which may ultimately affect the results. Standardisation of SEPs also makes it easier to compare results, especially in the era of meta-analysis. A Standard Operating Procedure (SOP) is a step-by-step guide to performing a SEP. A specific task of the PIGWEB project, which has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770, is to develop SOPs for methods to collect common biological samples (blood, faeces, urine, saliva) and measurements (carcass and body composition) that also take into account the 3R principles. A general template, including materials, equipment, prerequisites and a description of the SEP was created and filled in by the partners to collect the SEPs used in the different PRIs. Five working groups in different areas then exchanged and defined the best practices for a SEP, with consideration of the 3Rs and a focus on promoting animal welfare. The use of common SOPs within and across PRIs has many advantages and provides a solid basis for obtaining high quality data, promoting animal welfare standards and improving research collaborations.

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Theatre 5

Streamlining pig microbiome studies: Improving efficiency through reproducibility and comparability

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The microbiome plays a critical role in animal health and productivity, with the porcine microbiome being a key marker of pig health and farm productivity. Despite its importance, variations in technical and methodological approaches don't allow the comparability and reproducibility of microbiome studies in pigs. Addressing this challenge, our project introduces a pioneering effort to standardize microbiome research in farm animals, using pigs as a model. We aim to develop a standardized working protocol complemented by documentation guidelines. This is expected to enable the comparison of microbiome data across diverse laboratories, facilitating rapid conclusions on the practical relevance and application of findings to enhance livestock welfare, health, and performance and support sustainable agricultural practices. Around 2886 publications were screened to identify relevant protocols from sampling to bioinformatics analysis. Metadata of 461 publications met the eligibility criteria of being in vivo studies and possessing sequencing data that is publicly available. To create a template for metadata collection, information regarding sample collection, storage, DNA extraction, sequencing platforms and strategies, quality control and clustering tools, taxonomic annotation tools and databases, and functional annotation tools and databases were collected. Our findings highlight the need for standardized protocols to ensure comparability and reproducibility in pig microbiome research, leading to more efficient research outcomes.

How to score diarrhoea in piglets? Fecal consistency scoring at piglet or pen level

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In order to evaluate dietary or management treatments on intestinal health of newly weaned piglets, fecal consistency scoring is often applied either at pen or at piglet level. A critical evaluation of the methodology is crucial to ensure reliable results and to identify the best method depending on the research question. In this study, we evaluated 1) the reliability of scoring at pen versus at animal level, 2) the variation in scoring between observers. In total, 28 pens with 5 piglets per pen were scored by the same five observers on d7 and d11 after weaning. Pen scores are based on what is visible on the floor surface. For the individual scores, fecal samples are taken in a transparent jar. Fecal consistency was scored on a tagged visual analog scale (tVAS) from 0 (no diarrhoea) to 100 (severe diarrhoea), with tags at every 20th unit. Each tag contained a picture as well as words describing the fecal excrement, such as hard/dry (0), normal (20), softer (40), yoghurt-like (60), watery/yellowish (80) and severely watery. A score equal or higher than 60 was considered as diarrhoea. Average score at individual piglet level was 34, ranging from 0 to 100. Observers scored between 11 to 36 animals out of 244 observations as having diarrhoea (>60). Average score at pen level was 31, ranging from 2 to 83. 0 to 8 out of 58 pen observations were considered as having diarrhoea. In 9 pens, at least 2 individual piglets were scored as having diarrhoea, but only 3 of these pens were indicated by at least one observer as such when scoring at pen level (Avg: 55±12). In 14 pens, 1 individual piglet showed diarrhoea, which was reflected in the score at pen level of 6 pens (Avg: 50±7). Finally, there were 25 pens in which none of the piglets showed diarrhoea, while 2 pens were scored >60 by at least 1 observer (Avg: 42±6). Overall, we observed a notable disparity in scores assigned by different observers at the pen level, contrasting with a closer alignment observed at the individual animal level. An overview of the ICC values of the two methods and how the methodology used affects the power/sample size will be presented.

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PIGWEB Map of European research infrastructures working on pig experimentation for farm applications

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PIGWEB aims to build a long-lasting European Pig Research Community, and one of its main objectives has been the creation of a map of Research Infrastructures (RIs) in Europe. The interactive map (map.pigweb.eu) based on information collected using an online survey, developing a comprehensive vision of existing RIs, their location and characteristics (e.g. installations, facilities, expertise on). Currently, 51 infrastructures are available on the map, 14 are infrastructures of PIGWEB partners, and 37 from other research organizations. PIGWEB Map is a user-friendly tool, where users of RI can find institutions with necessary facilities and expertise that may not be available in their organization. RIs can be found on the map through selecting the region of interest, to look for information through the filters (i.e., country, area of expertise, type of facilities, or partner institutions). Participation to be part of the map is voluntary and allows providers of RIs to be more visible, attract users and establish partnerships establishment and strengthen connections in the pig research community. To be part of the map, contact pigwebmap@irta.cat. PIGWEB received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

The strengths, weaknesses, and missing techniques or equipment of Research Infrastructures, assessed from a user's perspective

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The European Commission indicated that “Research Infrastructures (RI) are key elements and play a key role in the European research setting”. They contribute to the advancement of knowledge, attract young people to science, structure the scientific community, and are key in a movement towards “Open”, interconnected, and data-driven science. Nevertheless, member states remain central in the development and financing of RI. Livestock research faces the challenge that its RI can be very specialized, are expensive to operate and livestock research is challenged by society. One of the activities of the PIGWEB project is to exchange with the research community about the future needs for pig RI and share the outcome with European and national stakeholder. During this session of the EAAP meeting, a SWOT (strengths, weaknesses, opportunities, and threats) analysis will be carried out. In addition, an online survey will be held among users of RI. The interactive session and survey will be the basis to develop a set recommendation, which will be shared and discussed in next year's EAAP meeting after which it will be communicated to stakeholders. PIGWEB received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101004770.

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Poster 9

Is the ABCD standard appropriate for comprehensive metadata description for pig data?

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Despite growing awareness of the importance of reusing research data in animal science and complying with the FAIR principles, there is still a lack of suitable and comprehensive metadata standards for phenotypic data in pigs and other livestock species. Improving metadata description, including using standardized vocabulary, i.e., ontologies, plays a pivotal role in making data FAIR. Currently, phenotypic data is commonly shared through data repositories (i.e., Zenodo). The metadata typically provided is limited in comprehensiveness, primarily focusing on publication details rather than the intrinsic characteristics of the data. As part of PIGWEB and in collaboration with NFDI4Biodiversity, we studied and assessed established and comprehensive standards for their suitability in pig data. The Access to Biological Collections Data (ABCD) standard, created by the Botanical Garden and Botanical Museum in Berlin, is a possibly suitable basis for creating more comprehensive machine-readable metadata. We present our current status and possible extensions of the ABCD standard, and offer potential solutions to use the standard within the pig community. This could be seen as a starting point for demonstrating how richer metadata could be shared within the PIGWEB community and might serve as a template for other livestock species. Applying a more extensive standard will ensure that the data collected is described more FAIRly.

Antibiotics consumption in animal production in Europe – on the path to 2030 goals

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In the last decade, increasing effort for reducing the consumption of antimicrobials in livestock farming has been motivated by social and scientific communities demand, leading to significant legislative alterations. This occurs in the context of finding strategies to address antimicrobial resistance which have risen to alerting levels. The European Union (EU) has set objectives regarding the decrease of the use of antimicrobials. It is expected that EU Member States would reduce the sales down to 59.2 mg/PCU (in which PCU is Population Correction Unit) until 2030. In this communication, we delved into the official information of the antibiotic sales for all EU countries in the 2010-2022 timeframe. A statistical and multivariate data analysis, centered on the 2022's European Surveillance of Veterinary Antimicrobial Consumption (ESVAC) report on sales of veterinary antimicrobial agents, was performed to explore overall trends and identify the groups of countries and respective different rates towards the defined objective. Relation between national animal production profiles, calculated based on data extracted from the ESVAC interactive database (in PCU), and the antimicrobials sales in the corresponding countries (in mg/PCU) is observed. Also, difficulties regarding data collection and data uniformization throughout the countries are identified and discussed.

Dry-off practices on dairy farms as a tool for the reduction of antimicrobial use and improvement of cow health and welfare

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The increasing level of antimicrobial resistance harms the health and welfare of dairy cows, as well as the profitability and sustainability of farms, being a public health concern. The dry-off period is crucial as most antibiotics for mastitis prevention are used in this production cycle period. Intramammary administration of antimicrobials to all cows at drying-off is the most widely used approach for treating intramammary infections and reducing the risk of new ones in the following lactation. This study aimed to provide an overview of opportunities to limit antimicrobial use in Italian farms during the dry-off period and identify areas for future interventions. The survey comprised 20 closed-ended questions gathering info on herd composition, management, and housing and was conducted on 810 dairy farms in Northeast Italy. A key question was used to categorize the farms based on the percentage of animals treated with antimicrobials at the dry-off. Other questions were addressed to identify the main decision-making strategy at the dry-off time and farmers' opinions on mastitis issues. Farms using a selective approach to the transition to dry-off were more likely (44%) to implement a change in feeding regimen and reduce the number of milkings. Regarding changes in feeding regimes, the most common, with 85% of respondents, was a change to a forage diet to reduce milk production. When farmers were asked if mastitis was related to their dry-off management, more than 80% replied negatively, identifying as factors causing mastitis: cow cleanliness (34%), overcrowding (16%) and inadequate barns (15%). Therefore, it is very important to work on improving the farmers' education so that good farming practices, combined with proper field diagnostics, can improve the udder health of dairy cows.

Treated and untreated cows housed side by side in tie-stalls and their respective risk of harboring *E. coli* resistant to antimicrobials

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Parenteral antimicrobial treatment results in the excretion of antimicrobial-resistant bacteria. Dairy cows are commonly housed side by side in tie-stalls and frequently receive antimicrobial treatment. However, studies investigating treated cows as source of colonization of neighboring cows with resistant bacteria are scarce. Antimicrobial resistance (AMR) in cows (treated and untreated) in tie-stalls was investigated to assess their respective risks of carrying resistant bacteria. Furthermore, we analyzed associations of farm management with AMR. For isolation of indicator *E. coli*, rectal swab samples were taken. Cows were sampled depending on treatment history and location within barn (cow A: recently treated parenterally; cow B: untreated, next to cow A; cow C: untreated, at distance to treated cows). Susceptibility was tested by microdilution. Associations of AMR, treatments and management were analyzed using generalized mixed-effects logistic models. Susceptibility data on 571 isolates (131 farms) were obtained. No significant difference in prevalence of resistant *E. coli* was observed between cows B and C. Untreated cows had lower odds of carrying resistant *E. coli* than treated cows (B: OR 0.44, $P < 0.001$; C: OR 0.54, $P = 0.007$). Non-pansusceptibility (NS) of isolates was associated with antimicrobial treatment (1 treatment: OR 2.27, $P = 0.001$; ≥ 2 : OR 1.88, $P = 0.038$). Not using manure on forage crops was associated with NS (OR 2.01, $P = 0.004$). For daily practice, results of this study do not provide evidence for the need to separate treated cows from others during treatment in tie-stalls.

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Theatre 4

Evaluating the effect of motivational interviewing and farmer training on effectively delivering the UK levy board mastitis control plan

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Mastitis is a costly disease to the global dairy industry. Challenges like antimicrobial resistance and animal welfare demand a proactive approach to mastitis control. The Agricultural and Horticultural Development Board Mastitis Control Plan (MCP) offers an evidence-based route to mastitis control. While it is an established and proven industry tool, further development of compliance strategies to better understand the barriers to implementation of MCP diagnosis and solutions would benefit the industry. This pilot study investigated 2 social science approaches to motivating MCP implementation: 1) Motivational Interviewing (MI) techniques, 2) Engagement with the practical farm-based skills training delivered by veterinary surgeons (VS) to farmers, known as 'Farmskills' (FS). 20 MCP VS were recruited. MCPs were delivered by VS grouped into 4 social science categories (MI, FS, MI + FS) and Control. VS received training via an MI taster delivered from Sheffield Hallam University (SHU). For FS group, training was delivered via XLVets to farmers/managers and the MCP VS. Progress in clinical mastitis and bulk milk somatic cell count performance were measured after 12 months. Groups were compared by ANOVA. Compliance with MCP recommendations between farmers and VS was scored by self-assessment. Mastitis conversations between VS and farmers were coded by an independent coder at SHU for MI credentials using the coding instrument. No significant differences were identified between groups. This study, however, provides a platform for future work targeting veterinary communication training as VS deliver few MI skills in practice.

Predicting impaired postpartum health of dairy cows with prepartum blood serum profiles

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In this study we evaluated the value of prepartum clinical-chemical indicators to identify dairy cows at risk for impaired postpartum health. 180 cows from four farms were scored on 19 health aspects, once prepartum and twice weekly after calving until six weeks postpartum. Blood samples were taken once prepartum, in week one and week five after calving to determine concentrations of 13 serum variables. Per serum variable, a cut-off value was set to determine the desired range of each variable. Per cow, a Total Deficit Score (TDS) was calculated as integral indicator of postpartum health based on all clinical scores and postpartum results for serum variables. Cows were divided into three categories: low TDS (most healthy), high TDS (impaired health) or culled (during the study period). Univariate analyses were performed to evaluate if prepartum serum variables could serve as indicator for impaired postpartum health. The odds of having prepartum total protein (OR=8.2) and haptoglobin (OR=20.3) concentrations above, and prepartum magnesium (OR=6.9) concentrations below the cut-off value were larger for culled cows than healthy cows (all $P \leq 0.01$). Next, a multivariate analysis was performed to obtain a mixed model with an optimal subset of predictors. Higher parity, higher total protein concentrations, and lower prepartum magnesium and albumin concentrations were associated with impaired postpartum health (sensitivity: 15%, specificity: 97%). In conclusion, single prepartum serum variables could be used to identify cows at risk for impaired postpartum health, but the added value of a multivariable predictive model appeared to be limited at individual cow level.

Effects of supplementation with vitamin E or plant extracts on redox and immune status in early lactating dairy cows

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Early lactation is a period for oxidative stress and inflammation, which can lead to cell damage in dairy cows. Our objective was to investigate the influence of dietary supplementation of vitamin E or plant extracts, on redox and immune status of dairy cows. Forty-five Holstein cows, including 23 primiparous cows, were classified into 3 groups and followed from 3 weeks before to 12 weeks after calving. The 15 cows of the control group were fed an unsupplemented diet, the 16 cows in the vitamin E group received 3,000 IU/d 3 weeks before and 1,000 IU/d for 12 weeks after calving, and the 14 cows in the plant extract group received 10 g/d for 12 weeks after calving. Plasma redox status were analysed by spectrophotometry and HPLC methods. Plasma cytokines were analysed after an ex vivo challenge of whole-blood cells with heat-killed *Escherichia coli*. Blood neutrophils were analysed by flow cytometry to measure their production of reactive oxygen species under ex vivo stimulation. Data were analysed with a mixed model including supplementation, parity, week. Plasma α -tocopherol was higher in vitamin E group than in the other groups. Milk yield, plasma H₂O₂, plasma antioxidant capacity and plasma glutathione peroxidase activity were not influenced by the supplementations. Erythrocyte glutathione peroxidase activity tended to be higher and plasma malondialdehyde tended to be lower in vitamin E group than in the other groups. Vitamin E and plant extract groups had lower plasma concentrations of cytokines: CCL4 for plant extract group, IL-6 as a tendency for vitamin E group, and IL-8 for both antioxidant treatments. Both supplemented groups had lower MHCII⁺ neutrophil percentage, and lower ROS production than the control group. Our study showed that antioxidant nutritional strategies in healthy cows can affect the redox and immune status and may improve antioxidant capacities and reduce the inflammatory response.

Navigating antimicrobial stewardship in dairy farming: A cross-contextual study of dairy veterinarians' perspectives in Alberta, Canada and Northern Italy

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Antimicrobial resistance (AMR) poses a global health challenge with implications for humans, animals, and the environment. This study aimed to provide a cross-contextual understanding of veterinarians' role in antimicrobial stewardship (AMS) and their perspectives on current policy in Alberta, Canada, and Northern Italy. Ethnographic data were collected and revealed that dairy veterinarians in Alberta recognized a global responsibility for industry-wide AMS. Trade-off in the use of Critically Important Antimicrobials, and the role of the supply management system in antimicrobial prescribing practices were acknowledged by veterinarians in Alberta. The results demonstrated variability in defining AMS success, and reservations about adopting a "European" model. In Italy, the focus was on upstream drivers of AMU, emphasizing the function of prevention, animal welfare, and nutrition in AMS. Veterinarians perceived themselves as mediators between governmental policy, individual farm contexts, and farmers' belief systems. The results of the study underscore the importance of understanding the specific context prior to implementation of new antimicrobial policies and interventions.

Antimicrobial activity of bacteriocin producing *Lactobacillus* spp. against multidrug resistant bacteria

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Antimicrobial resistance has resulted in limitations on the use of antibiotics. Bacteriocins are proteins or peptides produced by bacteria such as lactic acid bacteria (LAB) that have been considered as effective alternatives to commercial antibiotics. This study evaluated the antimicrobial potential of bacteriocin-producing bacteria against multidrug resistant (MDR) pathogens. The effect of different culture conditions was examined to maximise bacteriocin production of antimicrobial LAB cell free extracts (CFEs). Multi-drug resistant *E.coli* ATCC2471 was the primary pathogenic bacteria used in this investigation. *Lactobacillus* spp. strains were grown overnight in MRS broth. The cell free supernatant (CFS) was filtered using a 0.22µm syringe filter and antimicrobial activity was assessed by agar well diffusion in triplicate, a positive and negative control were included per plate. CFEs were taken at various time-points (day 1 – day 7), where pH, biomass & absorbance was quantified. Statistical analysis was performed using Minitab statistical software (version 20.4). One-way ANOVA with Fisher's least significant difference (LSD) was carried out to test for significant differences between means, at a 95% confidence level. CFEs of *Lactobacillus* spp. exhibited strong inhibitory activity against MDR *E.coli* (≥ 17 mm). Culture optimisations resulted in a $9 - 12.7 \pm 2.5\%$ increase in the zone of inhibition (ZOI) of LAB CFEs against MDR *E.coli*. This investigation demonstrates the potential of LAB to combat antibiotic-resistance, ultimately enabling and extending their use as novel feed ingredients for poultry and livestock.

The effect of feed additives on the impact of liver fluke infection in dairy cows

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An infection with the flatworm *Fasciola hepatica* (liver fluke) causes fascioliasis in ruminants and other grazing mammals, which leads to liver damage and other health issues. The development of anthelmintic resistance is a growing concern in the management of fasciolosis, that compels us to explore alternative methods of prevention and/or treatment. We studied the curative effect of natural feed additives on the impact on liver fluke infection in cattle. Currently, it is unknown whether and to what extent these agents indeed have a reducing effect on the infection in cattle. We examined the reducing effect on faecal egg counts using one of the two distinct boluses, one containing allicin (a compound found naturally in garlic) and the other comprising an extract from *Ferula asafoetida*. Naturally infected cows (200) at 10 different farms were selected based on a positive blood serum ELISA test, that was taken at least 6 weeks after the grazing season. Fifty cows received the allicin bolus and 50 cows received the *Ferula* bolus. At each farm a similar number of ELISA-positive cows served as negative controls. Individual faecal egg counts were scored prior to the treatment with the bolus and 4 weeks after the treatment. An additional ELISA was performed at 4 weeks after treatment. Faecal egg counts and second ELISA are collected in March 2024. Results will be analysed between March and May 2024 and presented during the congress. Keywords: *Fasciola Hepatica*, feed additives, ruminants, liver fluke

Effect of a natural solution on gastrointestinal parasitism modulation in heifers

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Gastrointestinal parasitism affects the productivity, health, and welfare of young ruminants, particularly during their first grazing season. The use of antiparasitic drugs has been criticized due to the increasing parasite resistance and adverse effects on the environment and digestive microbiota. There is a growing demand for alternatives, including natural plant-based solutions, to prevent and control internal parasitism in livestock. To test a new solution on *Ostertagia ostertagi* and *Eimeria bovis* infestation, a 56-day trial (D0 to D56) was conducted under controlled conditions with 16 heifers aged 9.1±0.7 months, divided into two homogeneous groups (control or supplemented) naturally infected with parasites. Strongyle fecal egg counts, serum pepsinogen, and hematocrit measurements did not differ between groups throughout the trial. After coproculture, larvae developed from supplemented animals were less motile than those from control animals ($P<0.01$), which could limit their pathogenicity. Lower concentrations of plasma lipopolysaccharides were measured in supplemented animals only at D56 ($P<0.01$), suggesting that the colonic epithelial barrier could be less impaired by *Eimeria bovis*. However, the number of *Eimeria bovis* oocysts excreted did not differ between groups after D0 ($P=0.19$). These results suggest that the tested solution could favorably modulate gastrointestinal parasitism in livestock. This dietary approach represents a promising way to reduce the use of chemical antiparasitic molecules. A complementary study will detail the impact of this solution on the intestinal ecosystem.

Host Defense Peptides-based nanoparticles for the treatment of Bovine Respiratory Disease

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Bovine Respiratory Disease (BRD) is a multi-factorial disease caused by bacteria and virus, combined with stress factors. In this study, we have tested the in vivo activity of a combination of recombinant Host Defense Peptides (Bac5, BNBD-1, BNBD-3 and LAP) embedded in Pluronic® F127 micelles (PM-HDPs) to evaluate if they can control BRD pathogens and disease progression in calves. A total of 42 crossbred unweaned male calves from different origins were selected with thoracic ultrasonography (TUS) score of 2. A control group (n=21 calves) and a treated group (n=21 calves) were treated intranasally with 5 mL of 20 mM HEPES or 20 µM PM:HDPs, respectively. Before and after treatments, TUS score was determined and nasal swab samples were collected for metagenomics analysis. Data obtained from metagenomic analyses and TUS score were analyzed by LR-Chisquare. Although changes in the evolution of TUS score were observed, no significant differences were observed between control or treated calves. Metagenomic results indicated significant differences between treated animals compared to Control ones. *Mycoplasma bovis* and *Pasteurella multocida* appeared in control animals on day 6 but they were undetectable in treated animals (p<0,05). These results indicated the clear potential of HDP-based nanoparticles to control the presence of BRD pathogens in calves' respiratory tract.

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Theatre 12

European Swine Influenza Network (ESFLU) for the benefit of One Health

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Despite the burdens caused by swine influenza A virus (swIAV), surveillance across Europe is scanty and fragmented, disease awareness is low, diagnostic protocols are not harmonized, standardised procedures are lacking and vaccination coverage is inconsistent. New swIAV variants constantly emerge and the virus remains persistent over the year in many pig farms. Recent human pandemics and the zoonotic potential of swine influenza highlights its risks for public health. A One Health approach to swine flu is therefore needed. An interdisciplinary expert network of over 150 experts from 30 countries (ESFLU, <http://www.swineflu.eu>, 2022-2026) was launched, funded by the European Union (COST CA21132). The objectives are: facilitate data sharing and analysis for swIAV surveillance; support pandemic preparedness; strengthen capability for swIAV detection and characterization; establish guidelines for swIAV management and control; promote dialog between stakeholders. First year's achievements are establishing the network, reviewing existing swIAV monitoring and surveillance programs, reviewing literature on vaccine efficacy, training to strengthen laboratory and analytic capacities, facilitating exchange programs, connecting to OFFLU (global flu network) and WHO and producing the first report of European circulating swIAV strains sequences.

The use of targeted selective treatment in lambs to reduce wormer resistance

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Wormer resistance is a major problem in sheep gastrointestinal nematodes, as such wormer use needs optimised. A pen-side tool called the 'Happy Factor' algorithm was created to help farmers target wormer use. It combines weight and grass availability to calculate minimum growth targets for individual lambs. The Smartsheep projects aim was to develop an accessible platform for the algorithm that combined a dashboard and pasture growth app on a cloud-based system. It was validated on 17 UK-wide sheep farms. A questionnaire was given to participating farmers to understand user-friendliness, barriers to uptake, and the benefits of using the platform. As part of the project, the study farmers supplied faecal samples for faecal egg counts (FEC), nematode species composition, and wormer efficacy. In total 1545 FECs were processed from 14 farms. Mean FEC pre-treatment was 274 epg. Various drug classes were used, 3-ML Clear most commonly with a 78-99% efficacy range. *T. circumcincta* was most prevalent pre-treatment and post-treatment varied per farm. Benefits identified were reduced reliance on wormer and slowing wormer resistance. Challenges identified were set up time, equipment cost and hardware compatibility. Overall, 79% of respondents would recommend the Happy Factor to farmers.

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Poster 14

Comparative analysis of antimicrobial usage in Swiss farms housing pigs exclusively or cattle alongside

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Antimicrobial resistance (AMR) is an increasing threat for human and animal health and imprudent antimicrobial usage (AMU) is a major cause for the development of AMR. The Swiss Federal Food Safety and Veterinary Office established the national reporting system IS ABV for monitoring and quantification of AMU in Switzerland. The objectives of this study were to analyze AMU in fattening pigs using IS ABV data. Differences in AMU were investigated between farms housing pigs exclusively and mixed farms housing pigs alongside cattle. AMU was calculated in total for all farms (n= 99) and specifically for the antimicrobial classes of penicillins and tetracyclines. Calculation was carried out using a treatment incidence (TI) based on Defined Daily Doses (DDD) by the European Medicines Agency, 2016. From January to October 2022 there were a total of 232 prescriptions in 99 farms, including 146 prescriptions in 49 pig farms and 86 prescriptions in 50 mixed farms. A total of 4'475 out of 48'848 fattening pigs (=9.16%) were treated. A total AMU of 0.28 DDD/pig/year (min: 0.00028; max: 30.7) for pig farms and of 0.09 DDD/pig/year (min: 0.00275; max: 4.96) for mixed farms was calculated. For penicillin the mean TI for pig farms and mixed farms was 1.11 DDD/pig/year and 0.40 DDD/pig/year, respectively. No significant differences between the types of farms were found concerning total AMU or any specific active substance. No association of specialization of the farmers on pigs on AMU was found in this study. Overall low AMU in combination with moderate sample size could have limited the detection of such effects.

Effect of dietary microbial muramidase supplementation on weaned pigs' performance, health status and intestinal microbiota

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Microbial muramidase (MM) has recently been introduced as a feed additive for swine. Its efficacy in improving growth performance and gut functionality has been extensively studied in broilers. However, only a few studies have reported its effect on pig performance and health, while none have focused on their intestinal microbiota. Therefore, the present study aims to fill this gap by analysing the effect of dietary MM on the growth performance, health status and faecal microbiota profile of weaned pigs. A total of 96 weaned pigs (28 ± 0.2 days of age) were weighed and divided into two homogeneous groups for their body weight (BW) and litter as follows: (1) Control (CTR): standard diet; (2) Treated (TRT): standard diet supplemented with MM at 65,000 LSU(F)/kg feed. Pigs had access to the experimental diets from day (d) 0 (at weaning) until d44. Antibiotic treatments, faecal score, BW and feed intake (FI) were recorded weekly. Faeces were collected at d9 and d28 for microbial analysis (16S analysis). Results showed that supplementing weaned pigs with dietary MM increased FI ($P=0.05$) and ADG ($P=0.02$) throughout the entire experimental period, resulting in up to 2.1 kg of BW ($P=0.05$) at d44 compared to the CTR group. Furthermore, the TRT diet improved the feed conversion ratio ($P=0.05$) from d7 to d14 and reduced antibiotic treatments ($P<0.01$) until d28 compared to the CTR diet. Diet did not influence faecal score and mortality. In conclusion, the results confirm the benefits of MM on weaned pigs' performance and health status. Further analysis will investigate whether MM has affected the intestinal microbial profile. Acknowledgement: Operation 16.02.01, project MELiOR DIET (ID:5404635), RDP of the Emilia Romagna Region.

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Poster 16

Addition effects of MGM-P, Quebracho Tannin Product, from weaning to finishing pig

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To find alternative antibiotics in the pig feed, before present study, we studied the series of different additive levels of MGM-P experiments, 0.2%, 0.3% 0.5% and 1.0% respectively. Above results showed the addition level of 0.5% MGM-P best affection to improve the growth performance and alleviating piglet diarrhea. To find MGM-P optimal addition concentration the experiment was conducted with two stages. First, from 21age weaning to 49day days old. And second, from age 49day to slaughter weight over 110kg. 24 piglets weaned at 21 days of age were divided into two groups. The treatment group added 0.75% of MGM-P (quebracho tannin product) to the basal diet, the control group added 0.01% Flavomycin to the basal diet. The treatment group significantly reduced diarrhea incidence without any negative effect to daily gain, average daily feed intake, and feed conversion ratio. Mushy feces occasionally observed in the control group, which easily self-healed in a short period, and no infectiousness was observed in the same pen. The pathogenic bacteria that can commonly cause diarrhea in piglets were not found. The results of relative quantitative PCR showed that the relative gene expression of F18, as well as *E. coli*, was not different in the feces samples between two groups. At the second stage, the control group only fed a basal diet without antibiotics, MGM-P did not show an improved growth promoting effect, altering the intestinal flora structure. But it showed a tendency to reduce some of the free amino acids of the meat.

Development of a multi-class and multi-residue methodology through UHPLC coupled with high-resolution Orbitrap mass spectrometry to detect veterinary antimicrobials in bovine milk

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Sensitive analytical techniques have become more and more important to detect antimicrobial residues in food, especially with the advancement of mass spectrometry as confirmatory technique. This study aimed to develop a multi-class methodology using UHPLC coupled with high-resolution Orbitrap mass spectrometry to detect and quantify potential antimicrobial residues in bovine raw milk. Notably, for β -lactams the method served as a multi-residue approach, detecting diverse penicillin and cephalosporins. The study revolved around the determination of the definitive methodology to reach satisfactory recoveries for the selected analytical standards (ampicillin, cefalonium, cloxacillin, danofloxacin, dimetridazole, erythromycin, nitrofurantoin, penicillin G, sulfadimethoxine, tetracycline, thiamphenicol) and its subsequent validation. The extraction process included the addition of acetonitrile, an extraction buffer followed by a cleaning step using QuEChERS, sample filtration, and concentration. A single injection detected both positively and negatively charged analytes in a 20 min chromatography run using full scan mass spectrometry and polarity switching ESI mode. Sequences were analysed and corrected through TraceFinder® 4.1. The methodology was subsequently applied for the analysis of potential presence of specific residues at distinct time points, in particular penicillin G and cefalonium deriving from mastitis and dry off treatments in dairy cattle. Considering the results, future improvements could be made to reduce matrix effect and employ more appropriate internal standards for each analyte.

Intramammary Platelet Lysate Preparation as a Dry-off Treatment Tool for Dairy Cows

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Reducing the use of antibiotics in dairy cattle, mainly for clinical mastitis treatment during lactation or dry-off prophylaxis, is essential for a One Health approach. In this study, we tested platelet lysate (PL) as an alternative to antibiotic administration at dry-off for its anti-inflammatory properties. We collected whole blood from donor cows on 17 farms in the Po Valley, Veneto region, Italy, and processed it into PL syringes for intramammary use (5 ml derived from 1.5×10^9 PLT/mL PL). We collected milk samples from 621 quarters before drying for microbial and somatic cell count (SCC) analysis as an indicator of udder health; the SCC value was then converted into a linear score (LS) to classify quarters as healthy ($LS \leq 4$), moderate LS ($4 < LS \leq 5$) and elevated LS ($LS < 5$). Healthy quarters were divided into three groups and treated with sealant only (SO), platelet lysate plus sealant (PLS) or antimicrobial plus sealant (AS). Quarters in the moderate and elevated LS groups were divided into two subgroups and treated with PLS or AS. After calving, milk samples were collected to detect new infections, and ΔLS was calculated to show changes in the health status of the quarter. The results show that in quarters with $LS < 4$, there was no significant difference ($P > 0.05$) in the variation of ΔLS over the dry period between treatments (SO, PLS and AS). For moderate and elevated LS quarters, there is no significant difference ($P > 0.05$) in ΔLS between PLS and AS. This study suggests that the use of PL could be an effective alternative to antibiotics at dry-off for risk quarters, reducing overall antibiotic consumption.

Lactic acid fermentation - an alternative for disinfection in case of an animal epidemic?

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In the context of animal epidemics, affected farmers are subjected to various regulatory obligations. The process of final disinfection includes the treatment of liquid animal by-products (slurry) and feed residues. Usually, the slurry can be used as organic fertilizer. Infectious slurry needs to be disinfected first. One of the two common methods is chemical disinfection. The other strategy involves long-term storage periods ranging from three to six months. Ultimately, there are some requirements, difficulties, and side effects to note at the application which have a significant economic impact on the farmers. The aim of the project is to find an improved method for disinfecting slurry and deactivate including pathogens. Finally, there should be a reduction of 99,99 % of pathogens within a period of less than three months at its best within four weeks. The established method is based on lactic acid fermentation. Lactic acid fermentation is activated by the addition of carbohydrates to the slurry, leading to the growth of lactobacilli, which produce lactic acid and consequently lower the pH value. Through acidification, pathogens should be inactivated. In best case, the remaining feed residuals can be used as carbohydrate source to enable the process. Advantages of the method are the utilization of infectious and non-infectious material, saving storage capacity and reduce the environmental impact. Furthermore, the material can be used as organic fertilizer earlier, and the risk of reinfection decreases due to the rapid reduction of pathogens. After successful laboratory-scale experiments, the investigation of the method is now scaled up to more realistic conditions. This research is funded by VDI/VDE/IT GmbH.

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Poster 20

Associations of Swiss national reporting system's antimicrobial use data and management practices in tie stall dairy farms

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Antimicrobial use is above target and requires reduction especially in dairy cattle. Although AMU in dairy farms is high, studies estimating AMU specifically in tie stall farms are scarce. The objectives of this study were to estimate AMU using the newly established Swiss national reporting system for AMU in livestock and to identify associated factors on Swiss tie stall dairy farms. We calculated treatment incidence (TI) by using the EMA's methodology and their DDD standards. Data on factors potentially associated with AMU were obtained through personal interviews with farm managers on 221 farms. Retrospectively, during a 1-year period, data on a total of 7619 treatments were extracted from the national database. Associations between management factors and TI were analyzed using a glm with gamma distribution. Mean TI was 5.46 DDD/cow-yr (\pm sd: 4.10 DDD/cow-yr). Intramammary treatment accounted for highest TI (3.24; \pm 3.16 DDD/cow-year); dry-cow therapy accounted for lowest TI (0.44; \pm 0.49 DDD/cow-yr). Five of the investigated management factors were significantly associated with TI. Organic production (-2.16; 95% confidence interval [CI] -3.53, -0.41) and herd size (-0.81; CI -1.21, -0.34) were negatively associated with TI. Specific cow breeds (Brown Swiss, Holst. Friesian: 1.56; CI 0.35, 2.72; 1.42; CI 0.02, 2.90, respectively; refe: others) and use of hygienic powders on the lying area (1.10; CI -0.004, 2.15) were positively associated with TI. The national reporting system is a valuable tool for AMU estimation.

Innovative approaches to assessing sustainability: A case study of sheep and beef systems at North Wyke Farm Platform

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The role of ruminant livestock in protein production is crucial, requiring strategies for sustainable intensification. Studies conducted at the North Wyke Farm Platform, a facility within the Global Farm Platform, from 2018 to 2023, provide insights into the sustainability of sheep and beef systems. Before implementing interventions, establishing an economic sustainability baseline among systems was essential. Practical recommendations were given for stocking rates and herbage production in grass-based systems. Innovative approaches were used to assess economic-environmental trade-offs, revealing a positive correlation between environmental performance and economic profitability. Methodological aspects were explored, uncovering potential underestimation of environmental impacts when using representative animal approaches. Studies on nitrous oxide emissions challenged traditional methods, highlighting the importance of evaluating environmental impacts at a system scale. Animal health and welfare studies contributed to understanding respiratory health in beef calves. Qualitative Behavior Assessment and other protocols were used to assess animal welfare. Metrics were developed to explore the use of early-life liveweight for predicting carcass quality and propose a framework for quantifying the informational value of industry-recommended KPIs. A nutritional analysis of beef demonstrated that temperate pasture-based beef can be considered a single commodity in terms of nutritional quality, challenging conventional mass-based carbon footprint approaches. Sensitivity analysis provided critical insights into the choice of metrics in sustainability analyses. A study comparing different grass-based silage diets showed minimal differences in enteric methane emissions from cattle over winter.

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Theatre 2

Choreographed interactions with cows on a demonstration farm to rethinking livestock subjectivity and Human-animal collaborations

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Livestock are more than just sources of food and opportunities for circularity; they are sensitive beings with which humans build relationship through labour. This study aims to rethinking the collaborative relationships between humans and animals in livestock farming. To achieve this, scientists and artists collaborated on a project involving choreographed interactions with cows. Dancing moments took place in the fields and in the barn in an experimental farm of INRAE, a member of the Global Farm Platform. Researchers and animal handlers observed the interactions then shared their perceptions and feelings with the dancer. We shed light together on the role of emotions, bodies, subtle knowledge, but also observation-contemplation in farming practices. Doing this, we highlighted the invisible working relationships between animals handler and animals. Furthermore, animal handlers emphasized the importance of observation and spending time with the animals, a practice once disregarded as unproductive, in order to understand the herd, their behaviour and their becoming. The invitation to dance with the cows provided us with an insight into the farmers' mindset, their labour, and their connection with their animals. The collection of various elements throughout the process, including testimonies, videos, and photos, resulted in the production of a sociological and anthropological analysis, as well as a documentary intended for both scientists and the general public.

Future sustainable grazing systems at the UCD Lyons Farm Long Term Grazing Platform

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Globally, livestock systems face multiple challenges including reducing negative environmental externalities, while meeting an increasing global demand for animal proteins, coupled with reduced resource accessibility, and increasing legislative requirements. Pasture based cattle systems upcycle human inedible biomass to produce high quality proteins and micronutrients. In Ireland, the 'Long Term Grazing Platform' (LTGP) at the UCD Lyons Farm, was developed to address the multifunctionality of pasture-based livestock production systems including GHG emissions, biodiversity, land and resource use, pasture resilience, water quality, animal performance, product quality and economic sustainability. Comprising 24 ha (12 x 2 ha hydrologically isolated paddocks) the LTGP supports three (8 ha each) experimental farmlets dedicated to a fully integrated, technically efficient, dairy calf to beef farm production system. Sward types include a perennial ryegrass monoculture (PRG); PRG plus white clover (PWC) and a six species sward consisting of 2 grasses (PRG and Timothy), 2 legumes (white clover and red clover) and 2 herbs (Chicory and Plantain; MSS). Since its inception in 2019, compared to the PRG system, the MSS system has produced 25% more ($P < 0.05$) herbage per ha using 60% less fertilizer nitrogen input. Compared to PRG, the MSS grazing animals have higher growth rates resulting in a 5-week reduction ($P < 0.05$) in slaughter age, a 14% reduction in the carbon footprint of each kg of beef produced and a 68% increase in net margin (profitability) per ha. The LTGP serves as a key national education and knowledge transfer resource, hosting multiple stakeholder groups, government Ministers and policy makers to inform the future of sustainable livestock production.

Impact of temperature on milk yield: a multiyear analysis from Harper Adams University Future Farm

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Climate change is a global issue with extreme weather such as heat waves impacting on livestock productivity, health, and welfare. Herein we analysed the impact of heat stress on milk yield and discuss potential mitigation strategies. We used milk production data from Harper Adams University Future Farm indoor dairy unit for 4 years (2019 to 2022). Meteorological data were obtained from Harper Adams University weather station and consisted of hourly temperature and relative humidity used to calculate Temperature Humidity Index THI. Data were analysed for mean milk yield per cow using a linear mixed effects models by month (range 28.7-30.3 L/cow) and THI, using year as a random effect. We report a significant overall effect of month on production ($F=10.8$, $P < 0.0001$) and THI ($F=6.7$, $P = 0.01$, Range, 26.6-71.6 °C) on milk yield. During warming months in the summer (May, June, and July) and in the Autumn cooling months (November and December), milk yield was also lower. The finding showed that heat and cold stress can impact milk yield. This pattern may become more prominent under climate change therefore the need to consider management intervention which may include breeding animals carrying slick gene with greater tolerance to heat stress, dietary strategies to reduce the production of metabolic heat, and engineered approaches to include buildings with fans alongside nature-based solutions.

Living labs concept towards GHG emissions reduction targets, growing market access and creating industry value in Australia

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Agriculture in Australia directly contributes to 14% of national greenhouse gas (GHG) emissions. The Zero Net Emissions Agriculture Cooperative Research Centre (ZNE-Ag CRC) was established in 2024 to build on 20+ years research in mitigation of GHG in Australia. It aims to coordinate industry, community and government actions in order to achieve lower GHG emissions while supporting the growth of agricultural industries in Australia. It will apply a model of 'Living labs and producer demonstration farms' and 'Lab-to-landscape' approaches to integrate research and innovative technologies for reducing GHG emissions and work with industry partners to co-develop projects to ensure research delivers practical and affordable solutions. In a coordinated network of producer demonstration sites representing major agroecological zones, results from laboratory and small-scale trials will be validated in commercial settings and GHG emissions reduction technologies will be scaled and run in combination (stacked), to evaluate impact on farm system emissions. At the same time, the demonstration of these technologies (including impact on profit) at a farming system scale, together with close co-operation from grower and producer groups will be a major route to adoption. Demonstration sites will provide a hub for sector-wide outreach and engagement that is both feasible and accessible to drive adoption of emission reduction activities, methods and initiatives from the CRC.

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Theatre 6

A grazing living lab in Northern Sweden a case study

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A grazing living lab (GLL) is an experimental and collaborative concept where on-farm practices related to grazing livestock and agricultural systems are demonstrated, evaluated and interpreted in a larger context. The vegetation season in northern Sweden is short and intensive, which calls for specific management procedures and use of novel technologies to achieve productivity. The GLL in our region serves as a platform for researchers, farmers, and other stakeholders to actively engage in the development of innovative practices in relation to improved use of grazed grass and enrichment of the landscape biodiversity. The GLL in Northern Sweden (Norr Nod) have selected a set of commercial beef, dairy and sheep "lighthouse farms" which have an inspiring pasture based animal production focusing on the grazing season. Among the identified farms, four are situated in Norrbotten and nine in Västerbotten counties, including Röbbäcksdalen, SLU's experimental dairy farm in Umeå. Our GLL hosts several activities including workshops, seminars, PhD courses, grazing experiments and study visits with focus on grazing sheep and cattle. From these, our presentation features a study conducted on two dairy farms, where we evaluated remote sense tools to predict pasture quality and estimation of potential enteric methane emissions during grazing. Pasture samples were taken for chemical and botanical composition to improve proximate models for estimating biomass production and grass quality. Several additional initiatives are on the horizon within the GLL, including efforts to extend the grazing season in the north and evaluation the use of virtual fencing technology for intensive and extensive grazing systems.

Katanning Research Farm Demonstration of Carbon Neutral by 2030 and Natural Capital Accounting

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Katanning Research Station is a 2000 hectare dryland grazing and grain cropping farm in south west Australia (Mediterranean climate) which is run as a commercial farming operation with 7000 head of Merino sheep. In 2020 we undertook a detailed carbon account and developed a practical plan to achieve carbon neutrality by 2030 as well as improve our farm's natural capital increasing the sustainability and resilience of the farming ecosystem into the future. Since this time we have achieved a reduction of 15% of our annual emissions, revegetated and rehabilitated 88ha of degraded saltland with suitable forage species and revegetated 25ha of waterways with native species of trees and shrubs for ecosystem revitalisation. We have made efficiency improvements in our sheep flock and pasture base by introducing new legume varieties in rotation with cereal grains, leading to lower supplementary feeding and better health. Currently we are planning our integrated shade, shelter and bio-energy plantings for 2025-28 to support microclimate and carbon opportunities. We expect to achieve 35% reduction by 2026. The Station runs key grazing, genetics, intake and methane studies but also plays an important role within the region for farmers to come and see successes or otherwise in implementing technologies and practices. We work on the principle that if we can or can't make it work in a commercial setting, you the farmers get to see that and provide opportunities for both farmers and scientists to work together to build a practical and profitable system that supports a better farming environment and quality food and fibre. See more at www.agric.wa.gov.au/carbon-farming/katanning-research-facility-carbon-footprint-assessment-and-strategy-achieve-carbon.

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Theatre 8

The living lab approach in the ecological transition of livestock farming: the case of the Abruzzo mountains

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The living lab approach in the ecological transition of livestock farming: the case of the Abruzzo mountains Since the 1960s, European agriculture has experienced dynamics of intensification, concentration, and specialisation, with climatic and environmental effects that are evident today and which have opened the reflection on the food system transformation processes and the ecological transition of livestock farming. High-altitude farming systems offer the opportunity to delocalise livestock activities, ensuring eco-systemic services and overcoming the competition between food and feed production. To move from ambition to action, technical, socio-economic and governance aspects must be considered, the management of which requires multi-actor processes aimed at creating new scenarios, convergent visions, and rapid processes of legitimised change. Living Labs (LL) represent a complex but potentially effective opportunity. The LL organised in the Abruzzo mountains, as part of the PNRR AGRITECH project, was carried out in June 2023 and consisted of company visits and Focus Groups (FG) coordinated by students and researchers, with the participation of local actors. The contents and operational proposals that emerged from the FG were presented at a final political discussion table with regional actors and institutions. The outcome of the LL was the launch of a possible national pilot project on sustainable livestock production in high-altitude territories, leveraging several tools, including community development, technological innovation, and the enhancement of livestock multifunctionality.

Living labs for assessing the overall sustainability of livestock systems through Life Cycle Assessment - SOLSTICE

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The Agritech Hub is an Italian Center for Technology in Agriculture, organised in 9 spokes: in the frame of Spoke 5 – Sustainable productivity and mitigation of environmental impact in livestock systems – a network of Living Labs (LL), known as SOLSTICE, has been established, covering a range of farming systems in different regions of Italy. The SOLSTICE initiative encompasses a diverse range of farmed animal species and categories, including dairy and meat cattle, buffaloes, goats and sheep, as well as pigs, rabbits, and poultry. Currently involving 31 stakeholders (farmers, associations, industries, policy makers, advisory services) and 6 universities (Padua, Bologna, Milan, Naples, Pisa, Tuscia), SOLSTICE activities focus on defining and transferring innovative management practices (feeding strategies, farm management, and manure management), along with implementing new technologies and monitoring systems in field conditions. The overarching objectives are to reduce greenhouse gas emissions (GHGe), improve animal health and welfare, and enhance overall farm efficiency. A simplified software application designed to assess the environmental performance of farms through Life Cycle Assessment is a pivotal deliverable of the SOLSTICE initiative. The challenge, but also the strength, of SOLSTICE is to apply the same systematic approach to completely different supply chains, encompassing both intensive and alternative livestock farming systems in highly productive as well as in marginal areas of Italy. The present work will showcase preliminary results from two case studies concentrated on enhancing the sustainability of the free-range poultry meat supply chain and applying tools for the monitoring of GHGe.

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Theatre 10

Innovative Tools for Assessment and Authentication of chicken meat, beef and dairy products'

QualiTies: a living lab approach

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This contribution presents preliminary results of the INTAQT project (EU Horizon 2020), which aims to perform an in- depth multi-criteria assessment of the relationship between animal husbandry systems and the quality of animal products. In specific, this research aims to identify and implement on-farm changes in the production processes (e.g. feeding regimes, outdoor access, herd management), which are expected to improve intrinsic quality traits of the products and/or sustainability traits of the farms. A participatory approach was used to establish farmer field-groups (living labs) representative of the different geographic regions and of the main production systems involved in the project. Each farmer field group involves 5 farms. The groups are established considering different husbandry systems according to a gradient of intensification (extensive vs intensive systems): 3 groups for dairy farms (Ireland, northern Italy and France); two groups for beef farms (Switzerland and northern Italy); two groups for poultry (France and Italy). The methodological approach is based on 5 steps: 1 -starting analytical phase: a critical analysis of trade-offs / synergies between sustainability and quality traits for each farm-field group; 2 – decision phase: development of practices to improve the identified synergies / mitigate trade-offs; 3 – implementation phase: implementation of practices for at least one year; 4 – concluding analytical phase: analysis and presentation of the effects of the practices during a last meeting in the farmer's groups; 5 -scientific data analysis and interpretation. Different innovative solutions are tested in each living lab. Some case studies will be presented to evidence the potential contribution given by living labs network to the improvement of food quality and sustainability.

The “Cheese Valley” Initiative: co-design innovations in the Pecorino Toscano DOP Value Chain

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The Cheese Valley Living Lab (LL), part of the AGROMIX project (Horizon 2020 N.862993) focuses on the Pecorino Toscano PDO value chain in southern Tuscany, Italy. The LL involves a multi-actor group comprising local dairy sheep farmers, advisors, the cheese factory board, policymakers, scientists, and retailers. The study area faces environmental challenges related to climate change such as extreme rainfall events, and summer droughts. To address these issues, farmers have been supported in adopting agroecological practices and agroforestry (AF). The initial SWOT analysis identified history and tradition as strength points, lack of innovation as weaknesses, opportunities in research and collaboration, and threats like uncertainty in production costs. A participatory mapping activity was conducted to understand AF knowledge gaps. On-farm experimental trials were conducted to collect data to improve knowledge about the actual implementation of AF systems. Poplar trees were planted on dairy sheep farms to increase fodder availability for lactating sheep in summer. The feeding trial, based on providing poplar stems and leaves to ewes, was carried out in 2022 and 2023, showing promising results. Furthermore, a study focused on rebranding the products was conducted in collaboration with a design institute. Challenges in the LL included rebuilding trust among stakeholders. Future steps include extending successful field trials to other farms and addressing the aging farmer population by training new farmers.

Co-designing livestock-based circular agri-food systems in developing countries rural areas: The CLiMiT living lab approach

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Many studies show that livestock within a circular agri-food systems framework can contribute to promote sustainability transitions in the context of developed countries. However, there is a lack of studies in rural areas of developing countries. CLiMiT project aims to develop an approach to facilitate the codesign and the implementation of livestock-based circular agri-food systems to promote sustainable territories in developing countries. Based on a Living Lab approach we first propose an interdisciplinary and participative agri-food systems territorial diagnosis starting from a biomass perspective. The biomass flows at farm and territory level and their governance are assessed through individual interviews and participatory workshops. Second, supported by multicriteria assessment methodologies (socio-economic and environmental), livestock-based circular agri-food system and the pathways to implement the transitions are co-designed with local stakeholders. Finally, via local NGO, we facilitate the implementation of the co-designed system and follow up in itinere the first obstacles for its operationalisation. The first phase of the project is set in two rural pilot areas located in Malagasy Central Highlands. The knowledge and tools mobilized will contribute to develop the CLiMiT approach that will be tested and improved in other developing countries during the second phase of the project

LivingLab - Effluents and coproducts of the livestock activity

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One of the priorities of the Innovation Agenda Terra Futura, of the Portuguese Ministry of Agriculture and Food, for the development of the Agrifood Sector, is Circular Agriculture. In this context it was approved a Living Lab (LL) on Effluents and coproducts of the livestock activity. The objectives of this Project are: 1) the implementation of a centralized LL with experimental and demonstration Units, spread all over the country, that answer to specific effluent and waste management issues; 2) to scale up the valorisation of livestock effluents and co-products of the livestock activities, as resources (TRL 6-7); 3) assessment of the impact of emerging solutions on the different processes to be developed; 4) assessment of substrate quality scenarios vs developed technologies vs valorisation of resulting co-products and respective impacts on the value chain. A multiactor approach, focused on the production and integrated management of the different flows generated at farm scale, foresees their use as secondary raw materials, recovering energy and nutrients, improving farm nutrient balances and promoting sustainable management. The main activities to achieve these objectives relate with the following routes: manure processing; feed and fertilizer replacement to develop a sustainable livestock production, bioenergy production at the farm, bioremediation by BSF larvae and biorefinery processes. This LL includes twelve partners from Research and Teaching, Agri Associations and Companies, as well as 3 entities of national and transnational cooperation. The LL is supported by PRR, PRR-C05-i03-I-000218.

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Universal bovine identification with depth cameras and deep metric learning to underpin the John Oldacre Centre dairy research platform

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The John Oldacre Centre for Dairy Welfare & Sustainability Research at Bristol Veterinary School is a new research platform based on intensive monitoring of our commercial dairy farm. A network of video cameras has been installed to capture and characterise activities and behaviours of each individual cow 24/7, which will be linked to health, welfare and sustainability. Real-time identification of individual cattle is an essential step towards achieving this goal as well as its translation to next-generation precision livestock farming. Previously we have demonstrated excellent results based on identifying the unique coat patterns of Holstein-Friesians (<https://doi.org/10.1016/j.compag.2021.106133>). Here, we present a novel deep-learning approach via a top-down depth camera perspective that instead leverages 3D cattle body shape only. We study the PointNet and Residual Neural Network architectures, which respectively ingest point clouds and depth maps of cattle, and compare their performance with the SOTA models that consume coat pattern images. Together, with this, we present the CowDepth2023 dataset, comprising colour-depth and point cloud tuples of 99 cows collected at the John Oldacre Centre over a 3 month period. Our findings suggest that depth-based ResNet and PointNet can accurately identify individuals, achieving $\approx 98\%$ and $\approx 99\%$ accuracy, respectively, on par with the coat pattern-based model. Our approach also avoids the need for network retraining to account for herd population changes by combining the network embeddings with the k-nearest neighbours algorithm. With our new approach we can now reliably identify animals using depth data alone, despite body shape variations across time, opening up video-based tracking to breeds that do not have distinctive coat patterns. The capability to track each animal in real-time in video over wide longitudinal time scales could lead to earlier detection of health issues, tailored interventions, and ultimately, improved overall welfare of the livestock.

Insights and experiences of two Living Labs in Tuscany (Italy)

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Living Lab (LL) is an innovative tool constituted by a multi-actor working group that develops, tests, and evaluates innovative solutions to solve specific problems of an agricultural sector. The LL Consorzio Tutela Pecorino Toscano DOP (LL1) and Granducato (LL2) are part of Codecs (No. 101060179) and Re-Livestock (No. 101059609) Horizon 2020 projects. The LL1 is located in the South of Tuscany region (Italy) and deals with the sheep milk devoted to the production of Pecorino Toscano DOP cheese. The LL2 is located in the North of Tuscany and deals with the dairy cattle farms for fresh milk production belonging to the Granducato farmers' cooperative. The aim of LL1 was to analyze the best digital innovations to be implemented in the dairy sheep sector and to assess costs and benefits of farm digitization. The LL2 aimed to evaluate dairy farmers' knowledge and perception for innovative practices to reduce the environmental impact and to increase the resilience to climate change. In both LLs, local farmers, consultants, the cheese factory technicians (for LL1), Farmers' cooperative (for LL2), policymakers, researchers, and retailers were included. In both cases, the participatory approach allowed to identify the main issues of the sectors and the innovative solutions able to help the farmers to overcome the problems. Decision support systems implemented in mobile app, sensors for monitoring animal behavior and health status and other innovations related to animal feeding and management were discussed and classified for the level of interest by all the actors of LLs. In any case, technical assistance and economic support have been identified as limiting factors for the application of innovations.

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Climate Farm Demo: A European-wide network of pilot farmers implementing and demonstrating climate smart solutions for a carbon neutral Europe

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Climate-smart farming (CSF) is an approach to agriculture and livestock that helps farmers grow resilience to the impacts of climate change, reduce greenhouse gas emissions, improve carbon sequestration promoting overall agricultural productivity and food security. EU launched CLIMATE FARM DEMO a 7-year project coordinated by IDELE (France) will promote CSF practices in 27 EU Member States covering all European pedo-climate zones and agricultural sector. The project aims to accelerate the adoption of these practices by farmers and all actors of the AKIS to adapt agricultural production systems to climate change and to reach a carbon neutral agricultural sector by 2050, through the establishment of an EU network of 1.500 pilot demo farms (60% livestock, 25% arable crops, 15% horticulture) and Climate Farm Advisors (CFAs) using a multi-actors approach at European and national level. Innovative CSF solutions will be co-created in a 10 Living Labs spread across Europe and will be demonstrated across 4500 demo-events. A set of private and public rewarding mechanisms will be identified, proposed and demonstrated to the AKIS actors, thus incentivising the uptake of CSF solutions while ensuring sustainable business models. CREA will coordinate engagement with policymakers, co-lead the rewarding mechanism analysis and all WPs activities with the cooperation of Apoconerpo, AIA and CRPA. The activity was funded by Horizon Europe work programme of the European Commission.

FERTY-Feed fertilization crops and energy: an example of co-design for the implementation of climate smart solutions in Italy

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In agriculture the main source of greenhouse gas (GHG) emissions comes from animals and within livestock sector the greatest contribution is given by ruminants and pigs. Pig farming is also the main responsible of ammonia (NH₃) emissions that cause awful smells; harmful to animal's and farmer's health and welfare. Pigs are produced mainly in the northern of Italy, a geographical area characterized by an inadequate level of soil organic matter (SOM), due to high exploitation of the soil to increase agricultural production. For this reason it is necessary to develop new climate smart solutions through the application of an innovative process of co-creation, co-design and experimentation in real conditions in a limited geographical context. The aims of the Italian living lab (LL) "FERTY" are to reduce GHG and NH₃ emissions by developing new and more sustainable feed formulations with lower crude protein (CP) content characterized by a partial replacement of corn and soy with alternative and local crops (i.e. barley, sorghum, sunflower and pea protein); and to increase the SOM using digestate, crop rotations and conservative agriculture according to the principles of the "BiogasDoneRight®" model developed by Italian Biogas Consortium in the heavy pig production chain. CREA and CRPA, monitor and facilitator of the Italian LL, will coordinate and encourage the engagement with all stakeholders (pig farmers, policymakers, feed producers, agrifood industry, agricultural machinery manufacturers), the co-creation process, the reflexivity and learning among participants, and improve decision-making. The activity was funded by Horizon Europe work programme of the European Commission under grant agreement n. 101060212.

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Steps for sustainable livestock: our multidisciplinary global farm platform journey

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The Global Farm Platform initiative (www.globalfarmplatform.org), established in 2014, is a network of research farms and institute members working collaboratively to enhance the sustainability of ruminant livestock systems through the development of transformational regional solutions to global challenges and promote their adoption. This multidisciplinary international network provides a unique combination of research and practice for diverse ruminant production systems in various cultural, socioeconomic, and climatic zones. Our steps for sustainable livestock include: 1) Feeding animals less human food, focusing on forage-based systems and reducing reliance on supplementary feeds; several research farms (RFs) are exploring sustainable practices, such as the Kenian "Tumbukiza" method and increased plant diversity in grazing systems, to enhance resilience and productivity. 2) Raising regionally appropriate animals adapted to local conditions; RFs are engaged in crossbreeding and selecting animals for resilience to climate change challenges, ensuring they can thrive in specific environments. 3) Keeping animals healthy; RFs employ various approaches, including technology like sensors and video cameras for monitoring animal health, as well as vaccine development to combat specific diseases. 4) Smart supplements; RFs explore options like spontaneous vegetation and innovative legumes to enrich soil quality and provide nutrition. 5) Focusing on quality over quantity in food production; RFs are implementing measures such as CT scanning for carcass confirmation and assessing the nutritional value and carbon footprint of forage-based beef systems. 6) Tailoring practices to local culture; promoting the transfer of research outcomes to stakeholders and farming communities. This includes initiatives to transform agriculture for environmental and economic benefits. 7) Tracking costs and benefits; involves assessing the environmental and economic impacts of sustainable grazing systems, utilising Life Cycle Assessment approaches, and collating databases for comprehensive analysis. 8) Studying best practices; involves the Global Farm Platform initiative's network of research farms globally. The initiative aims to optimise livestock use in various regions, considering local resources, breeds, and feedstuffs.

Transformative rumen microbiome research for on-farm implications

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In many nations, ruminant protein is an integral component of the human diets and its production contributes to economic development and poverty eradication. It is the forestomach known as the rumen that enables ruminants to thrive on grass pastures and digest fibrous plant materials that are unsuitable for most other livestock species. The rumen harbors a complex – symbiotic microbiome, which degrades plant cell walls and ferments sugars to the volatile fatty acids that provide energy to host. Advanced sequencing and omics technologies have shown that the gut microbiome and cattle genome interact to directly influence cattle productivity and health. We propose for a future “microbiome solutions” living lab initiative for cattle, that will collect biological samples (rumen and feces) and performance data on farms (both dairy and beef cattle) to generate data in support of precision supplements choice, targeted molecular breeding and feeding, silage processing as well as mitigation of environmental negative impact (methane and manure). Different microbiome solutions (bovine derived direct fed microbes, microbial enzymes, postbiotics) will be implemented to target feed efficiency, and methane mitigation traits. This initiative will collaborate with existing living labs in Canada, dairy and beef producers and Agriculture and Agri-Food Canada and University researchers. The “microbiome solutions” living lab aims to demonstrate how transformative animal microbiome research can support a collaborative framework that benefits Canada’s beef and dairy industries. Implementation of novel microbiome solutions could contribute to the sustainability of cattle production.

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Owned vs borrowed data: lessons learned from data collection on commercial dairy farms

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The Swedish University of Agricultural Sciences (SLU) has invested in two data collection infrastructures for livestock: SLU Basreg which collects data from the university’s three research farms, and SLU Gigacow which collects data from a network of commercial dairy farms collaborating with the university. Advanced commercial dairy farming equipment makes up much of the data collection capacity in both infrastructures, with the difference lying in the freedom of experimentation and access to the data generating environment provided by farms owned by SLU. Access to the sensors and systems generating data has also been important in the validation and understanding of data generated by SLU Gigacow. In many research projects the work on SLU research farms can form the nucleus for further studies followed by larger studies and validation in a commercial context by SLU Gigacow which also provides higher statistical power in genetics research or methods trials due to the greater size of the network. Through investing in Basreg and Gigacow, SLU has also become an important partner in the digital transformation of Swedish dairy farming helping both to develop new equipment and the means for data exchange. In the next phase of expansion, integration with commercial platforms such as Agronod and the Swedish node in the International Dairy Data Exchange Network managed by Växa will enable access to even larger cohorts of data. These integrations with commercial farms through the Gigacow network and new data sharing networks within the industry greatly reduce the cost per animal when conducting research. These multiple layers of research infrastructures will to a degree overlap in terms of the data collected and will interact in a manner similar to the phases of medical trials. Understanding the opportunities, limitations and investment requirements for each layer is crucial for the development of living labs in livestock farming. SLU is providing researchers with access to hundreds of animals for experimentation in Basreg, thousands of animals for validation studies in Gigacow, and soon the long term monitoring of tens of thousands of animals through data exchange networks.

Promoting rotational stocking in beef cattle pastures: insights from the Alberta AgriSystems Living Lab
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The use of rotational stocking in grassland may play an important role in improving soil C sequestration. One of the goals of the Alberta AgriSystems Living Lab (AALL) is to quantify soil C and greenhouse gas emission changes after implementation of rotational stocking, as well as estimate the impacts on the vegetation. Currently, 12 ranches distributed throughout Alberta have engaged with the AALL in a 5-yr co-development project, which started in 2023. The AALL has gained popularity and more producers are willing to joining in 2024. The producers involved have implemented or increased the intensity level of the rotational stocking. At each of these sites, ten GPSed marked plots were placed in representative paddocks within the whole pasture. Each individual plot measures 10 x 10 m. Within each plot, a 1 x 1 m permanent quadrat was placed in the center to estimate plant richness and diversity. In six out of the ten plots, exclusion cages were placed to estimate forage biomass, biomass removal, and nutritive value. Biomass removal is quantified by harvesting the forage inside the cage and in an outside-paired area two meters apart after grazing. Forage samples are divided into grass and forbs, and analyzed for crude protein, neutral and acid detergent fiber, and lignin. Within each of the ten plots, eight shallow (0-15 cm) soil cores were collected and composited for C, N, organic matter, texture, pH and salinity analysis. Deep soil cores will be collected in each plot at 90-cm depth in the Spring of 2024. These cores will be divided into four different depth increments and analyzed for C and N. Additional soil samples will be taken at the last year of the study, whereas vegetation measurements are being taken twice or more each year, depending on grazing management. This ongoing project aims to quantify the impact of grazing management adapted to each producer's reality, rather than mandating a fixed schedule or intensity. With the increasing participation of ranchers and farmers, the gradient of management levels expands, enhancing our understanding of the benefits of rotational stocking in the province and facilitating more efficient knowledge transfer.

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University of Wisconsin-Platteville (USA) Living Laboratory / Global Farm Platform

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Dairy farmers of the Driftless Area (DA) in the Upper Mississippi River Basin (UMRB) are faced with an array of climatic, agronomic, edaphic, and water quality related challenges. Here we outline our long-term grazing and cropland research as part of the Global Farm Platform (GFP) at the UW-Platteville Pioneer Farm, a living lab in the UMRB. This research compares conventional dairy production system common to the Midwest region of the United States, i.e., corn-on-corn (*Zea mays* L.) for four years, followed by alfalfa (*Medicago sativa*) for three years, with no cover crops with two alternative dairy production systems, (i) Soil Health Management (SHM) with no-till, cover crops, and application of a novel manure-based nutrient rich-stable product; and (ii) Management Intensive Grazing (MiG), rotational grazing on pastures established with diverse forage-legume mix. Meteorological, edaphic, hydrologic, and agronomic data are collected and analyzed at regular frequencies. Going forward, the experiment will continue to serve as a much-needed living laboratory for adaptive agricultural research in the UMRB.

Application of insects in poultry diets

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The increasing human population, evolving dietary patterns, and escalating competition between food and feed production underscore an imperative to develop innovative and sustainable food production systems. Among livestock sectors, poultry production has gained attention due to its notable environmental and economic advantages. Consequently, there is a pressing need to refine and innovate along the poultry value chain to enhance its sustainability. A pivotal challenge lies in transitioning towards sustainable protein sources in poultry feed. In this context, insects emerge as promising candidates. Species like the black soldier fly (*Hermetia illucens*) and yellow mealworm (*Tenebrio molitor*) have been identified as versatile mini-livestock for feed production. These insects possess a remarkable ability to convert low-quality organic waste into valuable body mass rich in high-quality protein and fat. When focusing on amino acid (AA) profiles, comparable values have been reported for insect sources and soybean meal. Regarding animal feed production, it is essential that insects fulfil the requirements of the target animal. In poultry diets, most limiting AAs are methionine, lysine, tryptophan, and threonine. According to the essential AA index for broilers, black soldier fly and yellow mealworm larvae exceed the required quantities of essential AA. Also the high concentration of medium-chain fatty acids, and especially lauric acid, in black soldier fly larvae fat is remarkable. Insect larvae are noted for their potential to enhance poultry health and welfare due to the presence of bioactive compounds such as chitin, lauric acid, and antimicrobial peptides, coupled with poultry's natural inclination towards them. This presentation aims to discuss recent research promising findings on various black soldier fly products and yellow mealworm as components of broiler and laying hen feed. However, further investigation is warranted to determine optimal inclusion levels, defining the nutritional and health-promoting characteristics of insect products, and evaluate their efficacy across diverse poultry production systems. Addressing remaining research gaps is critical for realizing the full potential of insect-based proteins in enhancing the sustainability and resilience of poultry production chains.

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Theatre 2

Application of less processed insect protein in broiler pelleted feed

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Eliminating drying when processing products into alternative protein ingredients for broilers reduces costs and environmental impact. However, increased moisture can negatively affect pellet quality and feed intake. Recently, black soldier fly larvae meal (BSFM) and oil (BSFO) received attention as alternative ingredients in broiler feed. Yet, processing demands energy and can impact the nutritional availability and functionality of insect products. This study aims to evaluate the effect of including PureeX[©] (Protix, the Netherlands), an insect product with 72% moisture and 11% crude protein, in the pelleted broiler feed. There were 3 dietary treatments and 8 replicates. Day-old male Hubbard JA757 broilers were assigned to 24 pens (19 birds/pen). From day one, birds were fed either a control diet or one of the 2 experimental diet programs. All diets were isocaloric and with the same digestible content of essential amino acids in each feeding phase. The experimental diets consisted of a basal diet with soybean meal (SBM) as the main protein source. To which either 5% PureeX[©], or 0.8% BSFM and 0.6% BSFO and 3.5% water were added, providing an equivalent amount of protein, oil and moisture as 5% PureeX[©]. Depending on the feeding phase, the control diet contained 6-10% more SBM than the two experimental treatments. Experimental diets contained 30% higher moisture in the starter and 20% higher moisture in other feeding phases, associated with higher pellet hardness. Overall, including 5% PureeX[©] or the mixture of 0.8% BSFM/0.6% BSFO and 3.5% water increased the feed intake by about 2%, which resulted in an increased feed conversion ratio (FCR). The negative effects were balanced out by calculating FCR based on dry matter. Results suggest potential for including less-processed insect products and reducing SBM in pelleted broiler feed without negative effects on growth performance.

Meat quality in broiler chickens fed a mixture of *Hermetia illucens* and *Tenebrio molitor* meals
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Hermetia illucens (HI) and *Tenebrio molitor* (TM) meals have widely been used in broiler chickens, but their mixture has never been tested. This study investigated the effects of HI and TM meals – alone and as mixture (1:1) – on meat quality of broiler chickens. A total of 420 1-day-old male broiler chicks were allotted to 7 diets (6 pens/diet, 10 birds/pen): C (control), HI5 (5% HI), HI10 (10% HI), TM5 (5% TM), TM10 (10% TM), MIX5 (5% HI-TM mixture), and MIX10 (10% HI-TM mixture). At slaughtering, breasts and thighs were collected for physico-chemical analyses. Thigh pH was higher in C and MIX5 than TM10 and MIX10, while higher breast cooking losses were observed in HI10 than C, HI5 and MIX5 ($P < 0.001$). Higher breast dry matter was observed in HI5 than TM5 and HI10, with the same broilers showing greater breast crude protein than TM5 ($P < 0.001$). Higher breast ash was identified in C and HI5 than TM5 and HI10, while HI10 displayed the lowest thigh crude protein ($P < 0.05$). In relation to breast fatty acid profile, the highest lauric acid was observed in HI10, with the same broilers showing higher myristic acid than C ($P < 0.01$). The highest palmitic, palmitoleic and oleic acids were identified in TM10, while the same broilers displayed the lowest linoleic acid ($P < 0.001$). Higher arachidic acid was observed in TM10 than TM5, while HI10 showed higher arachidonic acid than C, TM5, TM10 and MIX5 ($P < 0.05$). The lowest alpha-linolenic acid was identified in TM10, while the same broilers displayed higher SFA than C, HI5 and TM5 ($P < 0.05$). The highest and lowest MUFA and PUFA, respectively, were observed in TM10, along with the lowest PUFA/SFA ratio and n-3 and n-6 ($P < 0.01$). In conclusion, the use of HI meal at low inclusion levels (5%) may be preferable for preserving meat quality in broiler chickens.

Fecal Metabolome of Chicken Fed Black Soldier Fly and Yellow Mealworm-Based Diets
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Insect-based protein can be a sustainable alternative to soy in poultry nutrition. However, beyond their macronutrient value insect meals can also contain bioactive compounds such as antimicrobial peptides and medium-chain fatty acids. We hypothesized that dietary inclusion of black soldier fly larvae meal (HI), yellow mealworm meal (TM) or a combination of both might induce functional alterations in the gut microbiome of chicken. We fed diets containing 5% HI, 10% HI, 5% TM, 10% TM, 5% HI+TM and 10% HI+TM to Ross 308 chickens from the age of 0 to 38 days. Treatments were replicated in 6 pens with 10 chickens each. Two chickens per pen were sacrificed for cecal content sampling and their metabolome profiles were determined by the targeted LC-MS-based TMIC MEGA metabolomics assay (The Metabolomics Innovation Centre, Edmonton, Canada). The metabolite concentration data were subjected to principal component analysis (PCA) and ANOVA. A total of 601 metabolites were effectively quantified and the PCA did not reveal any significant clustering of the metabolite profiles according to the treatments. The ANOVA detected 9 significantly affected metabolites ($FDR < 0.5$) by the treatments: 3 amino acid derivatives, 2 acyl-carnitines, 2 complex lipids, 1 sugar, and 1 organic acid. Most significantly, fecal concentration of dopamine was increased in the TM-fed groups in a dose-dependent manner. In conclusion, dietary inclusion of insect meals altered the fecal metabolome, which can be attributed to feed-derived compounds and microbial metabolites. Gut microbiome-associated changes might help elucidate the impact of insect-based diets on poultry physiology and growth performance.

Black soldier fly meal inclusion in broiler and turkey diets: effects on growth performances, meat quality and health

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For several years, feed manufacturers have been searching for new protein sources to respond to the growth of the sector and to the challenges of sustainable development. Several studies were carried out during the NextGenProteins project (EU Horizon 2020 – 862704) with the aim of assessing the growth performances of poultry and fish when fed emerging protein sources, including insects. For poultry, experimental dose-response trials were conducted to evaluate the effects of partial substitution of soybean meal with black soldier fly meal (BSFM) on traits related to productivity, welfare, metabolism, and product quality. Then, in-field validation trials were carried out to confirm the outcomes of the dose-response studies. The dose-response trial showed that the inclusion of 9% BSFM leads to similar feed conversion efficiency compared to a soybean-based diet, while 18% worsened productive performance and body weight uniformity. At both inclusion levels, no adverse effects on welfare and meat quality traits were observed. For the in-field trials, feeds containing 9 and 5% BSFM were provided to broilers (growing and finishing) and to female turkeys (from 64 to 105 days), respectively. Overall, BSFM-fed broilers achieved similar performances than those receiving a commercial diet. Turkeys fed diets with 5% BSFM presented better performance than control birds (body weight: 10,173 vs. 10,057 g/bird; feed conversion ratio: 2.127 vs. 2.141; respectively, $P < 0.05$). Gut microbiota of turkeys was similar between treatments while an increase in butyrate level in the ceca has been observed in the BSFM group, which could be linked to gut health conditions. The meat quality parameters did not show any difference between treatments except for a slight reduction of yellowness. Overall, the outcomes of the NextGenProteins project allowed us to conclude that BSFM has great potential to be used in poultry feeding.

Full-fat silkworm-based diets for laying quails: impact on egg fatty acid profile

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The study tested different full-fat silkworm chrysalis (*Bombyx mori*) meal (SWM) inclusion levels into laying quails (*Coturnix japonica*) diet to evaluate the effects on the egg fatty acid (FA) profile. A total of 240 63-day-old female quails were randomly housed in battery cages (12 cages/treatment, 5 quails/cage), and fed for 8 weeks with one of the following diets: a control diet (C), and three experimental diets incorporating either the 4%, 8%, or 12% SWM (SWM4, SWM8, and SWM12, respectively). During the 8th week, seven eggs/cage were collected and homogenised to one pooled sample (12 pooled samples/treatment). On samples, the FA analysis was performed. The inclusion of SWM positively affected the FA profile of eggs, as a constant increase in the proportion of n-3 FA and a decrease in that of n-6 FA was observed from C to SWM12 eggs ($P < 0.001$). These were attributable to the FA composition of SWM, rich in n-3 FA (29.7% of total FA). The trend described above determined a progressive improvement in the n-6/n-3 ratio which reached the value 2.36 in the SWM12 eggs vs 9.82 in the C ones ($P < 0.001$). Also, overall saturated FA, monounsaturated FA and polyunsaturated FA proportions were affected by the dietary treatments ($P < 0.001$), with results not always attributable to the sole SWM inclusion, but also the consequent modifications of other dietary ingredients. Overall, the present study demonstrated that full-fat SWM is an effective ingredient to further improve egg healthiness, by enhancing the n-3 proportion and reducing the n-6/n-3 ratio.

Evaluation of dietary inclusion of *Tenebrio molitor* larvae on gut microbiota modulation in growing pigs
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Insects in animal nutrition are considered a promising sustainable protein source. This study aimed to evaluate the partial replacement of soybean with *Tenebrio molitor* meal on performance, diet digestibility, blood metabolites and intestinal microbiota in growing pigs. A total of 14 animals (~80 days old) were allotted to two groups (7 pigs/group): control group (CON) was fed a commercial diet containing 4% of fermented soy protein concentrate, and treatment group (TM) was fed a basal diet with 5% of *T. molitor* larvae meal in replacement of fermented soy protein concentrate for 28 days. Pigs were weekly weighed, and faeces and rectal swabs were collected for analysing nitrogen digestibility with insoluble ash method and gut microbiota through 16S gene amplification and sequencing. Blood was sampled on day 28 for the titration of albumin, globulin, and urea using an autoanalyzer, and for the determination of interleukin-6 concentration with immunoenzymatic method. No differences were observed in zootechnical performance and protein digestibility for the entire trial. TM revealed a difference for the beta diversity index at day 14 suggesting a microbiota modulation after the inclusion of insects ($p < 0.05$; R_2 0.1418). No differences were observed for the concentration of albumin, globulin, urea, and interleukin-6 in serum of both groups after 28 days, indicating a good health status. In conclusion, *T. molitor* meal could be considered as valuable feed ingredient for reducing the use of conventional protein sources in animal nutrition.

Increasing inclusion levels of *Alphitobius diaperinus* meal in juvenile rainbow trout diets
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Although extensive literature exists on the impact of including insect meal in rainbow trout diets, *Alphitobius diaperinus* (AD) still has to be examined in this context. The aim of this study was to evaluate the inclusion of partially hydrolysed AD meal at 6% (AD25) and 12% (AD50) in comparison with a commercial diet (C) as partial replacement (25 and 50%) of fish meal in the diets of juvenile rainbow trout. A total of 360 fish (average weight: 6g) were distributed to 12 tanks (4 replicates/diet) and fed by a feeding rate set at 2.7-3.3% of the biomass weight. At the end of the trial, considered when the fish triplicated their body weight, the assessment of growth performance, including final weight and feed conversion ratio (FCR), was conducted. Additionally, 13 fish per tank were killed through over-anaesthesia and 4 fish were used to assess the Fulton index, while the remaining ones were freeze-dried to determine the whole-body composition (dry matter, crude protein, ether extract and ash content, % on dry matter). Data was analysed by One-Way ANOVA (SPSS V20.0.0). The C-fed fish showed greater final weight (49.9g) when compared to AD50 ($p < 0.05$), while AD25 displayed an intermediate value ($p > 0.05$). In contrast, the FCR was not influenced by AD meal inclusion (47.7 and 45.5g; $p > 0.05$). The Fulton index was lower in C fish when compared to those fed AD meal ($p < 0.01$). No differences were observed in terms of whole-body composition ($p > 0.05$). Given the outlined findings, the AD meal emerges as a promising candidate for substituting fish meal. However, additional studies will be necessary to assess the maximum inclusion level.

Black soldier fly meal and dehydrated larvae inclusion in shrimp diets: effects on growth and health performances

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For several years, feed manufacturers are searching for new sources of proteins to respond to the growth of the sector and the challenges of sustainable development. At the same time, more than 30% of the world agricultural production is wasted. Black soldier flies (*Hermetia illucens* – BSF) make it possible to valorize food waste and represent an adapted resource to feed shrimps because it is natural, safe, sustainably produced and has a good nutritional quality. The objective of two studies carried out in Belgium in 2020 and 2021 and one study carried out in Ecuador in 2023 was to evaluate the performances of shrimps (*Penaeus vannamei*) when fishmeal (FM) is replaced by BSF meal in comparison to a conventional feed (CTRL). In the trials, a BSF meal produced in France or in Ecuador is incorporated into feeds at different inclusion rates as replacement material for FM (from 33 to 100% of replacement). These feeds have been used to feed shrimps from 0.24g to 10.3g. In the first trial, the optimal inclusion level was 12.7% (66% of FM replacement), the specific growth rate (SGR) was significantly better than the CTRL for this group of shrimps (p-value = 0.042). An increase in average final weight up till 16.8% for this group could be observed after 28 days. At the end of a complete culture period this would result in a substantial increase in productivity. In the second trial all dietary treatment containing BSF meal showed improved performances (weight gain, SGR, feed conversion ratio and survival rate) compared to CTRL even if not statically significant. In the third trial, the best result was for the shrimps feed with dehydrated grinded larvae replacing of 50% of FM (10.6g ; not significant). Our experience and the literature suggest a positive impact of BSF meal on growth performance in shrimp. These trials ended with bacterial challenges which showed promising results that will be presented.

Nutrient uptake, growth performance, blood metabolites and bone properties of broilers fed a ration containing mineral-enriched black soldier fly larvae

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Black soldier fly larvae (BSFL) can accumulate minerals, including P and Ca, from sewage sludge recyclates (SSR), and may therefore be used as a protein- and mineral-enriched component in poultry rations. In this study, the effects of supplementing SSR mineral-enriched BSFL on broiler performance and health were investigated. BSFL were grown on a fly diet (L-FD) without or with supplementation of two different SSRs, either with 4% bio-char (L-BCH), or with 3.6% single-superphosphate (L-SSP). A total of 80 day-old chicks (Ross 308) was assigned to one of 4 groups each with 6 replicate pens. All birds were fed age-specific basal diets and either had no access to BSFL (CON) or received ~ 15% of the feed intake (FI) of the CON birds daily as defrosted BSFL (i.e. L-FD or L-BCH or L-SSP) during a 42-day fattening period. On average, the broilers in the three larval groups consumed all BSFL in 15 min. Inclusion of 15% mineral-enriched whole BSFL in broiler rations had no adverse effects on growth performance, nutrient intake, nutrient conversion efficiency, plasma metabolites and immunoglobulins ($P > 0.1$). Birds in the BSFL supply groups had higher serum Ca concentrations than CON birds ($P < 0.05$). L-BCH birds had lower serum P than those in the CON group ($P < 0.05$). Tibial characteristics and mineral status of the birds were not affected by larval supply ($P > 0.1$). Heavy metal intake of the birds was not affected by the BSFL supply ($P > 0.1$). In conclusion, mineral-enriched larvae in broiler diets had no effect on nutrient intake, growth performance and bone condition.

Black soldier fly oil in the diet of dairy cows: effects on fatty acids of rumen digesta

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Ruminant livestock pose environmental challenges due to a high dietary dependence on imported plant oils and meals (from palm and soybean), which are considered no longer sustainable. Their replacement by insect products may be an alternative, but scant data is available. In this 50-day trial, we studied the effects of replacing hydrogenated palm oil (HPO) with *Hermetia illucens* oil (HIO) in dairy cow diet on ruminal fatty acids (FA). Twenty-six Valdostana Red Pied cows fed hay ad libitum were divided into 2 balanced groups that received isonitrogenous and isoenergetic concentrates containing HPO or HIO at 3% as fed (162 g/head × day). Rumen fluid was collected at the beginning and end of the trial and analyzed for its FA profile by GC-FID. Data were subjected to analysis of covariance. The HIO diet increased ($P < 0.01$) the proportions of 12:0, 14:0 and branched-chain FA, and it decreased ($P < 0.001$) the proportion of 16:0 in digesta. A trend ($P < 0.10$) towards higher t11-18:1 and c9t11-18:2 in HIO treatment was also detected, with no deviation in biohydrogenation pathways. No differences were found in the concentration of 18:0 and dietary unsaturated FA between treatments ($P > 0.05$). Overall, data suggest that HIO may be included as a sustainable feed in cow diets without impairing ruminal FA profile. Acknowledgement: project CSI020P23 (Junta de Castilla y León and FEDER/EU).

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Poster 12

Different EP-BSF levels and their effect on connexins, serum reproductive hormones, cytokine expression and the laying performance of hen at different stages

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In this study, a total of 250 Hyline brown laying hen of 1.91 ± 0.12 kg acquired at 24-weeks old were employed to evaluate the effect of different expanded black soldier fly meal (EP-BSF) levels and their effect on connexins, serum reproductive hormones, cytokine expression and the laying performance of hen at different stages. In the experiment, basic feed (CON), was compared with basic feed + 3% 5:5 EP-BSF (5:5 EP-BSF3), basic feed + 6% 5:5 EP-BSF (5:5 EP-BSF6), basic feed + 9% 5:5 EP-BSF (5:5 EP-BSF9), and basic feed + 12% 5:5 EP-BSF (5:5 EP-BSF12). At the conclusion of the experiment, 5:5 EP-BSF3 had higher linear hen-day-egg-production similar to CON when compared with 5:5 EP-BSF9 and 5:5 EP-BSF12 at the peak phase. The albumin weight was higher in 5:5 EP-BSF3, 5:5 EP-BSF6, and 5:5 EP-BSF6 compared with 5:5 EP-BSF9 at the peak phase. There was no significant difference in performances such as body weight, average daily feed intake, average egg weight, egg mass, feed conversion ratio, haugh unit, yolk colour, yolk percentage, albumin percentage, albumin height, eggshell thickness and hardness. We concluded from our result that the EP-BSF meal has the ability to reduce the dependency on corn, soybean meal, wheat bran, and animal fat by showing results similar to the CON diets when supplemented at level 5:5 EP-BSF3 and 5:5 EP-BSF6 in laying hen's diet.

Application of artificial senses to evaluate the effects of vacuum packaging on the organoleptic profile of dehydrated *Tenebrio molitor* larvae

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Insects are gaining more interest as novel food, but packaging might be essential to maintain organoleptic properties and enhance consumer acceptance. Thus, we aimed to evaluate the effect of the vacuum packaging on the preservation of *Tenebrio molitor* larvae (TM) organoleptic properties over time. Insects were dried, bagged and placed in non-vacuum (C) and under-vacuum (UV) conditions and stored for one, three and six months. One dried sample (T0) was immediately analyzed. An artificial sensory platform composed of an electronic Eye, an electronic Nose and an electronic Tongue was used to evaluate the color, the volatile profile and the dissolved inorganic components, respectively. All the data were subjected to PCA analysis. The storage time led to a darkening of all samples, but the UV group showed a set of color codes (from 819 to 1092) closer to that of the T0 sample (from 1090 to 2166) compared to group C (from 545 to 802). Also, the electronic Nose showed an altering effect produced by time storage. Indeed, the T0 sample was indicated by the LY-type sensors, while the stored samples were characterized by the P and T-type sensors. The most intense responses were shown by the group C, highlighting greater protein and lipid degradation compared to the UV group, as confirmed with the electronic Tongue. Altogether, the results showed the usefulness of the vacuum packaging to store *Tenebrio molitor* dried larvae and the effectiveness of artificial senses as a support for choosing the best packaging.

Session 96

Theatre 1

Exploiting big data for small ruminants

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Big data will enable animal sciences to embark on information technology-driven discoveries to face daunting challenges such as adaptation to climate changes, mitigation of environmental impacts, developing epidemiological models on current and emerging diseases. Toward this goal, Livestock Environment Open Data project (www. leo-italy.eu) has created a linked open data, merging existing and novel information on Italian livestock. Data are collected routinely on 18,194 livestock farms. Currently the database contains more than 15 billion triplets (ID-Data-Measure) generated, among others, by 30,074,112 dairy and beef cattle, 6,242,945 sheep and 1,328,356 goats. Specifically, for small ruminants, up to now, LEO has contributed in developing an indicator for ewe udder health. Through collaboration with the Sebastien project, Leo has provided data essential for modeling and mapping the risk of BT vector-borne diseases, as well as for developing a model for predicting pasture quality and grazing. LEO represents an opportunity for a new generation of scientists to engage with big data in livestock and develop advanced solutions applicable worldwide.

Virtual fencing vs physical fencing - grazing behaviour of goats

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The modern fencing technology virtual fencing (VF) requires the animals to associate the acoustic signal and electric impulse in order to avoid chronic stress. So far, studies, mostly conducted with cattle, could not detect major deviations in the normal behaviour of virtually fenced cattle compared to physically fenced (PF) cattle. In this study, we tested the VF system 'Nofence' with 20 'Blobe' goats with offspring in two subsequent periods. Two groups of ten adult goats each were assigned to a treatment (VF or PF) which swapped in the second period. The VF group had a virtual fence at one side of the paddock. Both periods consisted of a 12-day schedule which was divided into five phases. The goats' behaviour was observed daily via scan sampling during the four-hour trial time. The analysis of the grazing behaviour showed a significant effect of the interaction of treatment x period x phase ($P=0.038$) with more grazing shown by the PF group in phases two, three and five of period two. Since there were no significant differences in period one, we could not ensure that the shorter grazing time of the VF group in period two was caused by the VF system or other circumstances like the amount of herbage on offer. Further research is necessary to determine whether VF is a goat-friendly technology or whether it could induce chronic stress and negatively influence the goats' grazing behaviour.

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Theatre 3

Animal counting in outdoors systems based on drones and Computer Vision: from a review to the presentation of a new framework

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Counting the number of animals in a herd and ensuring their presence is fundamental to animal monitoring. However, in sheep farms where flocks can consist of thousands of animals scattered across vast rangelands, manual counting is challenging, time-consuming and often inaccurate. The use of drones combine with computer vision algorithm offers a promising alternative to traditional counting methods. A systematic literature review (n=37 relevant papers) permitted to find varying methodologies used in studies related to both subjects. While certain models exhibit high accuracy rates (up to 98%) in detecting animals, there is still room for improvement as errors could significantly impact large herds. Moreover, around 95% of these studies involved surveying the entire field with a drone, taking photos of all areas, and then using deep learning to identify cows or sheep. However, this approach faces many limits due to the legal restrictions regarding drones flights, and the density of the vegetation on rangelands that make animals invisible from the sky. To address this challenge, we suggest a new framework: using drones to record videos of sheep passing through a narrow passageway to facilitate counting. We propose to combine drone footage with crowd detection algorithm which permits to count every animal even in densely flock. To make this new framework a reality, we commit to openly sharing our dataset and algorithms on an Open-Access Drone Data Analytics Library (DDAL) for the research community. By fostering collaborative work and leveraging data sharing opportunities, we believe this project can offering considerable assistance to the farmers responsible for managing herds.

Implementing an autodrafter-weighing scale for assessing the body weight changes and grouping dairy sheep and goats during lactation

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Monitoring body weight (BW) changes during lactation is a key aspect for dairy sheep and goats managed in large flocks under intensive conditions. Most dairy ewes and does are allocated in groups according to lambing date and fed ad libitum a unique diet despite their level of production. As a result, body reserves vary largely among animals and its assessment is the main criterion for adjusting the quality and feeding level of the diet. With this aim, a 2-ways autodrafter weighing scale (Prattley-Datamars, SW), using ID5000 scale indicator and XRP2 electronic low-frequency (LF, 134.2 kHz) reader from Tru-Test, was implemented in the Experimental Farm of the UAB for BW monitoring in a flock of 150 dairy ewes (Lacaune and Manchega) and a herd of 40 dairy does (Murciano-Granadina) identified by LF ceramic boluses (sheep, 22 g; goat, 65 g; Datamars) milked twice daily in a 2×12 milking parlour (DeLaval, SE). The system used Bluetooth and wi-fi for device connections and cloud software (Datamars). Milking data was collected using MM25SG milk meters and DelPro v.5 software (DeLaval). Ewes and does graze jointly (6 h/d) and were complemented indoors with a total mixed ratio ad libitum. Weighing was done weekly after the a.m. milking and before grazing from parturition to dry-off with minimal human intervention. On average, weighing throughput until 150 d in milk were 288 ± 16 ewes/h (83.6 ± 0.7 kg BW) and 224 ± 20 does/h (48.4 ± 0.5 kg BW). The throughput increased with weighing sessions, reaching 400 head/h after 15 sessions. Recorded BW data was compared to a target prediction model using the cloud software and used to allocate animals of each species in 2 groups according body reserves and milk yield, to which 2 feeding levels were applied to save concentrate. BW and average daily gain were also used as an early warning system for detecting welfare issues. In conclusion, both sheep and goats adapted rapidly to the autodrafter-weighing scale and its use was easily implemented on dairy small ruminant farm conditions. Acknowledgements: UE Program H2020 (Project TechCare #862050)

Session 96

Theatre 5

Predicting dry matter intake in sheep using greenhouse gas measurements from portable accumulation chambers

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In many countries around the globe, small ruminants graze in communal semi-natural pastures in mountain and forest areas where it is extremely challenging to obtain any measurement or indication of their dry matter intake (DMI). However, a group of traits closely related to DMI is the greenhouse gas (GHG) traits like methane (CH₄) and carbon dioxide (CO₂) production. With the increased use of portable accumulation chambers (PAC) and mobile PAC systems it is now feasible to record sheep for CH₄ and CO₂ production in remote areas. In this case it may be possible to predict DMI from GHG traits. Mature ewes in early gestation from the modern Norwegian White Sheep (NWS) (n = 20), and the landrace Old Norwegian Spæl (ONS) (n = 20) were included in the experiment. The sheep were housed in individual pens and fed grass silage ad libitum. Feed intake was recorded continuously for 19 days using BioControl feed bins, whilst GHG traits were recorded in PAC once per day. In total 760 concurrent observations of DMI, GHG and body weight were available. An artificial neural network was trained using a standard 80:20 split with both breeds in the training and validation, resulting in a high validation accuracy R₂ of 0.73. Encouragingly, in a subsequent trial with 10 independent sheep fed fresh cut grass for six weeks the validation accuracy only dropped to R₂ 0.72. Lin's CCC was high at 0.84 indicating there is potential to capture a large proportion of information in daily DMI in sheep using GHG and production traits.

Digital tools and technologies: Irish sheep stakeholder opinions before and after training

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Sm@RT, an EU Horizon 2020 funded project, aims to improve the understanding, awareness and uptake of digital tools and technologies available to small ruminant farmers. To facilitate this, each of the 8 participating countries (Estonia, France, Hungary, Ireland, Israel, Italy, Norway, UK) hosted training days on farms. Training days consisted of workshops where stakeholders had a demonstration and opportunity to use digital tools/technologies. In Ireland, 3 training days were held where stakeholders were able to evaluate and use a variety of tools including Electronic Identification (EID) readers, EID weighcrate with autosorter, flock managements apps, sheep conveyor, lambing cameras and weather stations. Prior to and after each individual demonstration, stakeholders completed a short survey on their opinion of the tool or technology. After training, some of the stakeholders' opinions changed due to a better understanding of how to use the tool and learning about the potential benefits for their farms. Training days provided an opportunity for stakeholders to have hands-on experience and improve their understanding of the potential benefits of technologies/tools available to small ruminant farmers.

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Theatre 7

Budget process linked to partial indicators in dairy sheep farm

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Is important to link partial indicators both to economic (revenues and cost calculated at the time of material stock variation) and cashflow (at time of cash inflows and outflows for the material payments). Budget process can be integrated to support medium-term decision making process. The aim of this work, (set within PNRR Agritech 5.2.6;CUP J83C21000300006) was to set up a method to plan economic and cashflow predictions, by integrating partial indicators and budgeting calculation in a dairy sheep farm (n=150 milking ewes, Sardinia-Italy). A budget from March to May 2024 was structured. This period is usually critical for planning reproduction efficiency and the next year productivity. Cashflows of purchased and self-produced feeds were considered paid at market value, at 60 days and at delivery respectively. Milk price was split in basal (0.90 €/l, paid the next month) and final price (+0.90 €/l, end of the year). Milk yield and other costs were estimated from historical farm records. Were calculated: Income Equal Feed Cost (IEFC, in l), economic and cashflow Income Over Feed Cost (IOFC). Other cost (such as utilities and labor) were added to calculate economic and cashflow break-even point (BEP) per flock and milking ewes. Milk yield was estimated declining from 2.0 to 1.7 l/d per ewe, thus 9300 l in March, 8100 l in April, an 7905 l in May. Estimated economic IOFC was 12,847 €, 10,813 €, 10,813 € in March, April and May respectively. Estimated cashflow IOFC was 3,475 €, 5,085 €, 3,648 € in March, April and May respectively. IEFC was on average 2139 l/month whereas economic BEP was 441l/month. Cashflow BEP per milking ewe in May was expected 1.74 l/d, higher than 0.4 l/d of predicted milk yield. IEFC gives a partial view of the farm whereas BEP is more comprehensive. The average difference between economic and cashflow IOFC per milking ewes was 1.58 €/d. Also the average difference between economic and cashflow BEP was 0.81 l/d. In summary, it is relevant to monitor in farm routine both aspects. To change the budget output in May technical decisions can be made (i.e.: more incomes or less cost) in order to do reduce the risk of a negative cashflow.

Is there any interaction between feed efficiency and resilience in dairy ewes subjected to underfeeding? 1) Feed intake and milk yield

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It is still uncertain whether selecting ewes for higher feed efficiency (FE) may compromise their resilience to cope with nutritional challenges. In this study, we investigated the relationship between both phenotypes by examining the response and recovery of DM intake (DMI) and milk yield (MY) to underfeeding in ewes divergent in FE. During a 32-d pre-challenge period, 40 Assaf ewes were fed a TMR ad libitum and monitored for daily DMI and MY, as well as twice-weekly milk composition and BW, to calculate voluntary intake and estimate FE (through Residual Feed Intake). Sixteen selected ewes were divided in 2 FE groups: higher (H-FE; n=8) and lower (L-FE; n=8) FE, and subjected to a challenge for 26 d, receiving only 70% of their voluntary intake. Then, they were re-fed the TMR ad libitum for 25 d (post-challenge). DMI and MY were recorded throughout the trial. Data were fitted to a linear model and analysed by repeated measures ANOVA. Results showed that differences in estimated FE were mainly due to a trend to greater DMI in L-FE ewes ($P < 0.10$), as no significant variation due to FE was observed in MY. Less efficient animals showed more negative slopes for DMI ($P < 0.001$), both in the pre- and post-challenge periods. Regarding MY, no differences between L- and H-FE were detected ($P > 0.10$). In both groups, MY dropped with restriction and initial levels were not recovered in the post-challenge ($P < 0.001$), in line with the lactation curve. These results do not provide clear evidence for a link between FE and the ability to cope with underfeeding, but more research is needed to ensure that higher FE would not compromise resilience. Acknowledgment: PID2020-113441RB-I00 and PRE2021-098235 (MCIN/AEI/Spain).

Environmental impact drivers of mixed dairy sheep farms: insights from a combined LCA and multivariate statistical analysis approach

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A cradle-to-gate LCA study on 13 dairy sheep mixed farms highlighted significant differences in agronomic practices and livestock farming techniques, in terms of environmental impact calculated as carbon footprint per liter of fat and protein corrected milk (FPCM). As regard the global warm potential (GWP), the results of the study put in evidence the importance of the nature of data collection, particularly as regard the variables associated to enteric methane production that accounted for over 52% of total methane emissions. Detailed collection of data about feeding regimen during farmers' interviews allowed to adopt CNCPS model equations and to better calculate biogenic GWP. Additionally, the results highlighted the importance of improving forage digestibility and reducing external concentrate purchases, which accounted for over 10% of emissions, to mitigate total GWP. Cluster analysis revealed two distinct farm typologies: (i) large surface, large size of flock, and high productivity; (ii) smaller farms with small size of flock and lower productivity. PCA analysis showed that feeding strategies and input use efficiency were associated to environmental impacts: high content of starch in the diet was positively associated with milk production and negatively with GWP, while the higher content of fiber in the diet was positively associated with GWP as well as fuel consumption.

The effect of feeding a calcium peroxide feed additive to mitigate enteric methane emissions in lambs

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Introduction: Methane is a natural product resulting from enteric fermentation in the rumen forestomach of ruminants. Specifically, 70-80% of methane is produced by rumen methanogens through conversion of carbon dioxide and hydrogen. Various peroxide-based additives have been screened in vitro and shown potential for application as methane mitigating feed additives. This study investigated the methane reducing potential of a calcium-peroxide (CP) based formulation in an early-life lamb study, following on from a previous ewe trial that determined the optimal inclusion rate. **Materials and Methods:** Seventy pregnant Lowland crossed ewes (35 ewes/group) expecting twin lambs were selected and fed concentrate pellets (dry matter (DM) basis) as follows: control (0.0%) and CP (7.5%) two weeks prior to lambing. At weaning, each of the twin lambs was assigned either the control or treatment pellet, resulting in four treatment groups: Control x Control (CC), Control x Treatment (CT), Treatment x Treatment (TT) and Treatment Control (TC). Grass silage was fed ad libitum to both ewes and lambs, whilst the amount of concentrate was periodically adjusted (25% DM basis). DM intake (DMI) was recorded daily, with body weight recorded weekly. Enteric methane emissions were measured using a GreenFeed unit. Response variables were analysed using REML, with treatment as a fixed effect (SPSS, 27.0). **Results:** Lambs supplemented with calcium peroxide (TT and CT) showed a lower methane yield ($p < 0.003$), across the duration of the trial than those in control groups (TC and CC). Lambs in the treatment groups showed on average 30% greater daily live weight gains than those in the control groups, however this was not significant ($p = 0.699$). **Conclusions:** The supplementation of calcium peroxide has shown great potential in increasing productivity and reducing methane emissions in lambs. Metagenomic sequencing of this data will elucidate changes occurring at the microbiome level between each treatment group.

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Theatre 11

Reproductive performance of a new cervical fix-timed insemination protocol in ewes based on three doses of prostaglandin F_{2α}

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Due to its rapid rate of metabolization, easiness of application, animal welfare impact, and cost, prostaglandin F_{2α} (PG) represents a simpler option than progestogens-eCG protocols (P4-eCG) for the reproductive management of sheep. Increasing the interval between two PG doses up to 14 to 16 days apart ("2PG" protocol) determined better reproductive outcomes after cervical fixed timed AI (FTAI), achieved conception rates comparable to P4-eCG, but lower conception rate than spontaneous estrus ewes. With the aims to improve the reproductive outcomes of 2PG, this experiment evaluated the effect of pre-sync ewes with another PG dose supplied seven days before the first dose of 2PG ("3PG" protocol). During the breeding season (31° S-57° W; Salto, Uruguay), 200 multiparous Merino ewes grazing native pastures were assigned to "2PG": two 15 days apart (Day -15 and 0) and FTAI at 56 h after Day 0 (n=99; Control) and "3PG" group: three PG doses (Day -22, -15 and 0) and FTAI at 68 h after Day 0 (n=101). Cervical FTAI was performed used a fresh semen pool from 12 Merino rams. Visual-physical state of cervical mucus at FTAI, fertility, prolificacy and fecundity rates were evaluated by trans-abdominal ultrasonography on Day 60. Differences between groups were analyzed using procedure for categorical variables of SAS. It was observed a protocol effect ($P < 0.05$) in fertility rate (64.6 and 77.2%), but not ($P > 0.05$) in cervical mucus grade (2.17 ± 0.09 and 2.14 ± 0.10), prolificacy (1.30 ± 0.06 and 1.19 ± 0.04), or fecundity (83.8 and 92.1), 2PG and 3PG protocols respectively. We concluded that the 3PG protocol with FTAI at 68 h significantly improve the fertility outcomes compared to ewes synchronized with the 2PG protocol.

Factors affecting differences in goat milk yield and composition between evening and morning samples

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In dairy cattle several studies investigated differences in milk yield and composition between evening and morning milking's. Similar studies in dairy goats are lacking and milk secretion differs between cattle and goat (merocrine in cow versus apocrine in goat). Identifying and understanding factors affecting these differences is critical for choosing between alternate or continued milk recording schemes. Therefore, the aim of this study was to quantify differences in goat milk yield and composition between morning and evening milk samples and investigate to what extent these differences are influenced by genetics, herd, milking interval and the temperature-humidity index (THI). In Dutch goat milk recording schemes milk yield and milk composition of both morning and evening are routinely recorded. In addition, electronic identification of goats during milk recording provides information on milking time which allows calculating milking intervals. Heat stress might affect differences between morning and evening milking's and therefore information from weather stations was used to calculate the THI. For the current study in total milk 108,362 morning and evening milk production records were available from 7874 different goats located on 7 farms. Records were collected from 2018 till 2022. Test day averages were 3.9 kg of milk, 4.21% fat, 3.52% protein and 4.28% lactose. Differences between evening and morning milking's were -0.07 kg milk, +0.47% fat, +0.01% protein and +0.02% lactose. Results will be presented on how these differences are affected by genetics, herd, milking interval and THI.

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Theatre 13

Polyphenols from *Sulla* (*Hedysarum coronarium*) affects rumen fluid microbiota of dairy sheep ewes

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The present study was part of the project Disolasulla financed by the Italian agriculture ministry. The experiment was aimed at studying the effect of polyphenols from dehydrated *sulla* (DS), and *sulla* hay (SH) on rumen fluid (RF) microbiota and composition of Massese dairy sheep. The trial was carried out on 12 animals with a 3 x 3 Latin Square experimental design. Diets tested were Control (C, corn grain, faba bean grain and alfalfa hay) or two diets where alfalfa hay was substituted with DS or SH. Taxonomic characterisation of bacterial and archaeal communities was performed by sequencing V3–V4 region of the 16S rRNA. Methane was measured by a Laser Methane Detector. Volatile fatty acids (VFA) content in rumen fluid were determined by HPLC-UV and total fatty acid composition by GC-FID. Diets affected the composition of microbial communities. With respect to C, RF from sheep fed DS contained greater percentage of *Anaerovibrio* spp., whereas RF from SH contained smaller percentage of *Butyrivibrio* spp. The modification of microbial communities affected also VFA production. Sheep fed DS diet produced less propionic acid (4.3 mg/g of rumen fluid vs 5.6 and 6.0 mg/g of rumen fluid for SH and C treatments, respectively). In tendency ($p = 0.163$), also smaller amount of acetic acid was produced from sheep fed DS. The DS diet also affected the production of isovaleric acid, probably due to changes in rumen protein metabolism. Feeding treatments didn't affect Archaea abundance in RF, however, the amount of methane produced from sheep fed DS diet was in tendency reduced ($p=0.155$). Finally, DS treatment also affected fatty acids biohydrogenation by inducing an accumulation in RF of dietary omega 3 fatty acids and vaccenic and rumenic acid originated by fatty acid biohydrogenation.

Development of a minisequencing integrated method to genotype PRNP gene in goats

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Scrapie is a transmissible spongiform encephalopathy (TSE) that has been present in goats and sheep for more than 300 years. It did not represent an issue until the appearance of bovine spongiform encephalopathy (BSE) and the consequent social alarm it created. Breeding of genetic resistant animals can be considered an effective tool to control this disease in both species. That is why, for more than 20 years, in sheep it has been carried out a breeding program linked to genetic selection of resistant genotypes to promote resistance in classical Scrapie control. In July 2017, EU requested a scientific opinion from EFSA, and it has been established the evidence of nine amino-acidic polymorphisms in PRNP gene sequence related to the resistance of goats to classic Scrapie (G127S, I142M, H143R, G145D, N146D, N146S, R154H, R211Q y Q222K). The Department of Genotype of Laboratorio Central de Veterinaria in Madrid, has developed a fast and safe method to genotype these nine nucleotide polymorphisms using "primer extension". The following primers were designed: G127S: 5' CAGCTGGAGCAGTGGTAGGG 3', I142M: 5' AGTGCCATGAGYAGGCCTCTTAT 3', H143R: 5' TGCCATGAYCAGGCCTCTTATRC 3', G145D: 5' GATAGTAACGGTC CTCATAGTCAYYG 3', N146D: 5' TTCAYGATAGTAACGGTCCTCATAGTCAY 3', N146: 5' TTCAYGATAGTAACGGTCCTCATAGTCA 3', R154H: 5' TGGGGTAACGGTACATGTTTTCA 3', R211Q: 5' GAAACTGACATCAAGATAATGGAGC 3' and Q222K: 5' GAGCAAATGTGCATCACCCAGTAC3'. In their design, it has been included the existence of different polymorphisms in the junction zone of primers of different Spanish breeds (Blanca Celtibérica, Florida, Guadarrama, Malagueña, Murciano-Granadina, Negra Serrana o Castiza, Payoya), which has shown an effective tool in all breeds analyzed. In the present work, we show the results obtained in the analysis of different breeds.

Session 96

Poster 15

Data driven Innovation in Sheep Sector: APPàre, a platform to map technical and environmental performances from life cycle assessment (LCA) and costing (LCC)

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Data-driven innovation is crucial for EU food systems, but agri-livestock systems still lack digital infrastructure, hindering innovation. The project PNRR e.INS "Ecosystem of Innovation for Next Generation Sardinia" engages local communities in sustainability challenges. Under the SPOKE03: "APPàre: smart and secure livestock farm Applications to boost data driven innovation along the food chain" aims at mitigating social impacts of the environmental crisis and promoting territorial inclusion. It aims to develop a platform promoting data-driven innovation along sheep chain in Sardinia and to sustainably intensify livestock farms, while increasing their productivity and profitability. The software and hardware infrastructure is under development to create graphical interfaces of input collection and output dashboards. The life cycle inventory was carried out with comprehensive list of farm inputs valorizing already existing databases from regional institutions or private stakeholders and farm gathered records. A set of algorithms has been developed to perform calculations using IPCC 2019 coefficients in a simplified LCA and LCC for impact estimations. It includes key indicators of: carbon footprint, energy consumption, CO₂ capture and sequestration, soil land use and fertility indicators, and ecosystem services for social and biodiversity contribution. Managerial indicators include flock profile, feed efficiency, milk and meat productivity, income over feed cost and income equal feed cost. The tool provides guidelines for the widespread application with a potential diffusion on 10 000 dairy sheep farms in Sardinia. A business intelligence informative system is under development to foster consultancy and exploitation of gathered data. The use of the information platform might facilitate: i) farm data recording, storage and analysis, promoting efficiency in the production chain; ii) favouring climate change adaptation; iii) formulation of regional policies for eco-innovation, enhancement of ecosystem services, and growth of the sheep supply chain.

Is there any interaction between feed efficiency and resilience in dairy ewes subjected to underfeeding? 2) Ruminal fermentation parameters

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Due to the importance of rumen fermentation for diet utilization and feed efficiency (FE), this study was conducted in ewes divergent in FE and subjected to a nutritional challenge to explore variations in ruminal fermentation parameters that may serve as proxies for a potential relationship between FE and resilience. For a 32-d pre-challenge period, ewes were fed a TMR (50:50 F:C) ad libitum and monitored to calculate their voluntary and residual feed intakes. Based on the latter index (RFI), 8 sheep with the highest (H-FE) and 8 with the lowest (L-FE) FE were selected to undergo a 26-d nutritional challenge, receiving only 70% of their voluntary feed intake. This restriction was followed by 25 days of ad libitum TMR re-feeding (post-challenge). At the end of the pre-challenge and challenge, and at 2 times of the post-challenge, ruminal fluid samples were collected via stomach probe for analysis of ammonia and volatile fatty acids (VFA). Data was subjected to repeated measurement analysis. Differences were only observed among periods (i.e., due to the nutritional challenge). Thus, during the restriction, ammonia and butyrate remained unaffected, whereas acetate was increased and propionate decreased. Surprisingly, ammonia values were higher in the post-challenge than those found in the pre-challenge period ($P < 0.05$). The rest of the parameters recovered to initial values after the challenge. No significant differences between L-FE and H-FE groups were observed in any of the studied parameters ($P > 0.10$), which would suggest that more and less efficient ewes would probably display a similar resilience in terms of rumen fermentation. Acknowledgment: PID2020-113441RB-I00 and PRE2021-098235 (MCIN/AEI/Spain).

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Poster 17

Using image analysis to estimate goats live weight

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This study aimed to predict the goat's live weight using a digital image analysis system. Body weight is one of the most informative traits, which provides information on growth, feed conversion, feed efficiency and disease. 110 Bravia goat breeds with 37.1 ± 7.8 kg body weight were studied. Measurements were performed directly in goats, and equivalent measurements were obtained by image analysis. A system consisting of a video camera and a frame built with two red lasers of 650 nm wavelength was used for image capture. From the images obtained and using the program ImageJ, 19 morphometric measurements were determined. All statistical analyses were performed using the JMP program. A descriptive analysis of data, correlation coefficients between the measurements obtained by image analysis and measurements from the goats and stepwise multiple regression equations to predict goat live weight were established. A significant correlation was obtained between actual measurements and measurements obtained by image analysis ($P < 0.01$, $r > 0.8$). It is found that the equation, which includes the four body measurements obtained by image analysis, explains 89.7% of the body weight variation. This equation was obtained after four steps, including body area, thoracic area, and withers and hip height measurements. We conclude that image analysis has the potential for practical application in predicting the live weight of the goats. However, in future work, other measurements should be considered so that models predicting live weight prove more accurate. This work was supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Is there any relationship between feed efficiency and milk fatty acid composition in dairy ewes subjected to underfeeding?

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A previous study found that a brief but acute underfeeding (only straw for 3 days) altered the milk fatty acid (FA) profile in dairy ewes regardless of their feed efficiency (FE). However, it raised the question of whether there could be a relationship between FE and FA profile in ewes exposed to a longer nutritional challenge. To answer it, lactating sheep of divergent FE were subjected to a nutritional challenge in which they were offered 70% of their voluntary intake for 26 days. Pre-challenge data were collected from 40 ewes over 32 days to calculate voluntary and residual feed intakes. Based on the latter (RFI), we selected 8 ewes with the highest (H-FE) and 8 with the lowest (L-FE) FE, and they were subjected to the challenge. Then, they were re-fed TMR ad libitum for 25 days (post-challenge period). Composite 3-day milk samples were collected at the end of the pre-challenge and challenge periods, and at 3 times of the post-challenge, and analysed by gas chromatography. Data underwent repeated measurement analysis. Regarding variation between FE groups, the H-FE ewes showed higher concentrations of butyric, oleic, and monounsaturated FAs (MUFA, $P < 0.05$), along with a trend toward higher conjugated linoleic acids (CLA) and lower saturated FAs (SFA, $P < 0.10$). These differences are compatible with a healthier milk FA profile for consumers. As for the effect of the challenge, SFA decreased with feed restriction while MUFA increased significantly, these changes persisting in the post-challenge samplings ($P < 0.001$) in both high and low efficient animals. Statistically significant FE group \times period interactions were barely detected and corresponded to only minor, a priori not relevant, FAs. Acknowledgements: PID2020-113441RB-I00 and PRE2021-098235 (MCIN/AEI/Spain).

Session 96

Poster 19

Assessing the feasibility of Near Infrared Spectroscopy for the rapid detection of Aflatoxin M1 in goat milk
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Chronic dietary exposure to aflatoxins poses a serious public health risk due to their carcinogenic potency. Aflatoxin M1 (AFM1) is detected in milk and dairy products from animals fed with aflatoxin B1 (AFB1) contaminated feed. As climate change contributes to the increasing presence of AFB1 in raw materials and feed, the prevalence of AFM1 is rising. This research work aimed to assess the feasibility of Near Infrared (NIR) spectroscopy as a rapid tool to detect the presence of AFM1 in milk samples. A total of 108 milk samples collected from 18 goats from an AFB1-exposure trial were used. Laboratory results revealed the presence of AFM1 in 60 of the samples, whereas 48 tested negative. Milk samples were oven-dried on a glass-fiber filter and analyzed in a DS2500 NIR instrument. Discriminant models to discern between AFM1-containing and non-AFM1-containing samples were developed using six different algorithms available in the WinISI IV software. Within each group, 90% of the samples were used to develop, and 10% to validate the models. The Discriminate by X-residuals algorithm provided the best results, with 85% of the positive and 95% of the negative samples correctly classified in calibration, and 60% and 83% in validation, respectively. Although PLS2 multivariate analysis had slightly more limited results, with an average of 76% correct classification, the validation showed that all the positive samples were correctly identified, and only 2 of the negative samples were classified as positive, thus achieving an overall classification success of 82%. According to these findings, it can be concluded that NIR spectroscopy seems to be a promising tool to rapidly detect the presence of AFM1 in milk samples.

Water footprint assessment of by-products based diet for sheep production in Tunisia: potential of low water use diets

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Water scarcity is among the main challenges that render sheep farming systems in drylands vulnerable. The water footprint (WF) indicator has been proposed as a metric to measure the impacts of livestock production on freshwater resources. This study aimed to assess and compare the water use of sheep-based by-products feeding systems in central Tunisia, using the Water Footprint Network method. The primary data were obtained from 46 sheep farms located in central Tunisia, with the selection criteria requiring that the diets contain at least 30% by-products. A multivariate analysis was performed to characterize the different feeding systems. The typology analysis came up with five based by-products sheep feeding systems that are: olive cake (OCS, n = 14), date palm waste (DWS, n = 8), wheat bran (WBS, n = 18) and tomato pomace (TPS, n = 6). The WF of olive cake, date palm waste, wheat bran and tomato pomace were 1283, 917, 1517, and 1687 m³/ton of biomass, respectively. The WF of sheep meat produced under the target feeding systems ranged from 7654 to 9056 l/kg live weight. The evaluation of WF of the four sheep by-products-based feeding systems figured out that sheep raised under the DWS had the lowest WF per ton of live animal, while the TPS system exhibited the greatest WF. Water used to grow feedstuffs for sheep production accounted for 96% of the total WF of sheep. The green WF accounted for more than 83% of the total WF in all the studied feeding systems. This study showed the importance of feeding ingredients in the determination of the WF of sheep meat. Our findings suggest that by-product-based diets can significantly reduce the overall WF of sheep meat (20%) compared to conventional feeding system in central Tunisia. In conclusion, the strategic use of agro-industry by-products as low water feed alternatives for ruminants represents an innovative solution to the challenges of water scarcity in Tunisia. Such practices align with the principles of the circular economy on conserving water and contributing to the resilience of sheep production against water scarcity.

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Poster 21

Effect of the starch source on body fat deposition, carcass traits and meat quality of culling Sarda dairy ewes
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A fattening program for culling dairy livestock at the end of productive career could be a valuable opportunity for replenish body reserves mobilized during lactation and improve carcass conformation and meat quality. The purpose of this study was to investigate the effect of starch sources (barley vs. corn) on the evolution of body fat deposition, carcass characteristics, and meat quality of Sarda dairy ewes. Forty ewes candidates to be culled (initial average \pm SEM; BCS: 2.58 \pm 0.20) were equally allocated to two dietary groups according with the BCS, and fed ad libitum diets containing different proportion of corn and barley: diet 1 (CORN group) with 25% of corn, 15% of barley, and 60% of wheat middling; diet 2 (BARL group) with 25% of barley, 15% of corn, and 60% of wheat middling. Ewes of both groups had also ad libitum access to straw and water for 4 weeks until slaughter. During the trial, BW and BCS data were recorded once a week, and the daily feed intake of the groups has been recorded weekly. At slaughter, carcass measurements, pH and colour traits were recorded from the left side of the carcass. Proximate composition, cooking and drip losses were evaluated using the Longissimus thoracis et lumborum (LTL) muscle. The dietary treatment had no effect on BCS, live BW and slaughter BW. The CORN group showed a higher (P=0.039) cold carcass yield and a tendency for higher rib fat thickness (25.13 vs. 20.83 \pm 1.28 mm; P=0.068) compared to BARL group. Dietary treatments did not affect pH, colour, cooking and drip losses. The LTL of CORN animals displayed a higher moisture (70.65 vs. 68.73 \pm 0.45%; P=0.032) and lower protein (19.05 vs. 20.74 \pm 0.39%; P=0.028) contents. Lipid content of the LTL was unaffected by the diet. In conclusion, the type of starch did not affect body fat deposition, carcass characteristics and meat quality of culling Sarda ewes. In order to have a better knowledge about the manipulation of the meat quality of dairy ewes culled at the end of their productive carrier further studies are needed. Research funded by VALPESA project (PSR Sardegna 2014–2020).

ARIES: Advanced Research on the Impact of Environment on Sheep

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The overarching goal of the ARIES project, funded by the Italian Ministry for Universities and Research (PRIN 2022), is to contribute with specific knowledge and innovative tools to the Italian dairy sheep industry for improving its efficiency, and sustainability while guarantying animal resilience. Since the need of novel indicators to assess udder health, within the ARIES project we evaluate the possibility to use differential somatic cell count (DSCC) determined by automated systems in flow cytometry in sheep. A total of 720 individual milk samples were collected from two Italian sheep breeds, the Comisana and the Massese. In a subsample of 119 animals balanced between the two breeds, the DSCC was analysed based: 1) on the combined use of staining techniques, digitalization of microscope images, and cell counting software, 2) on the use of Fossomatic™ 7DC, a flow cytometer already used for calculating somatic cell count (SCC) in sheep's milk and DSCC in bovine milk. The correlation between the two DSCC values was calculated to validate the use of Fossomatic™ 7DC for measuring DSCC in sheep's milk. The correlation was equal to 0.57 (p-value < 0.001) with differences among the two breeds (0.61 in the Comisana and 0.50 in the Massese). Further studies on bigger sample size will be necessary to further validate the use of Fossomatic™ 7DC for determining DSCC in sheep's milk, nevertheless the obtained correlation seems promising. The results are embedded within the research contract of Giorgia Stocco, co-financed by the European Union – NOP Research and Innovation 2014-2020 art. 24, par. 3, A) Law 30/12/2010, n. 240 and of the D.M. 10/08/2021 n. 1062.

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Theatre 1

Somatotropic axis of Holstein cows of diverging genetic origin under two pasture-based systems

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Multiparous Holstein cows of North American (HNA, N = 24, 640 kgBW) and New Zealand (HNZ, N = 24, 575 kgBW) genetic origin were selected to assess somatotrophic axis uncoupling. During the prepartum, cows were fed a total mixed ration (TMR, 33:33:34 concentrate, barley straw, corn silage, dry matter (DM) basis). At calving, cows were assigned to two feeding strategies (FS), MaxP: 27% concentrate and 73% grazed pasture + forage reserves DM basis and FixP: 33% grazed pasture and 67% TMR 55:45 forage to concentrate, DM basis. Milk yield was recorded daily, plasma samples were taken fortnightly from prepartum to 112 days postpartum (DPP) and liver biopsies were taken at -14, 14, 56 and 112 DPP. Plasma insulin and insulin like growth factor I (IGF-I) were measured with commercial kits and mRNA abundance of IGF-I receptor (IGFI-R) and growth hormone receptor 1A (GHR1A) were studied using real time PCR. Data were analyzed using a mixed model with repeated measures that included genetic origin (GEN), DPP, FS and their interactions as fixed effects. Milk yield was 5.3 kg/d greater in HNA than HNZ from 35 to 112 DPP (P < 0.0001). Plasma insulin and IGF-I levels were 12 and 32% lower in HNA than HNZ, respectively (P < 0.05), and the lowest values for both groups were observed during the transition period (P < 0.0001). Hepatic levels of IGFI-R and GHR1A mRNA increased at 112 DPP (P < 0.05) while HNZ tended to have 1.6-fold higher levels of these transcripts at 112 DPP vs. HNA (P = 0.09). No effect of FS was observed. Although cows of both genetic origins experienced uncoupling of the somatotrophic axis during early lactation, there was a tendency for HNZ cows to experience recoupling before HNA cows.

Phenomic data integration for the identification of biomarkers of subclinical intramammary infection in Holstein cattle

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In this study we aimed at integrating different layers of phenotypic traits, specifically serum metabolome and blood parameters, along with milk leucocytes subpopulations, to investigate their relationships upon subclinical intramammary infection (sIMI) and identify potential markers for the disease in Holstein cows. Bacteriological screening on 188 mid-lactating Holstein cows reared in one herd under similar conditions was made to select: i) culture-negative animals with no history of mastitis (Neg, n = 19) and; ii) culture-positive animals for mastitis causing pathogens, that displayed no clinical signs of disease (sIMI, n = 21). Host-related traits included serum metabolomic profile measured with 1H-NMR (MET), oxidation (OXI; e.g., total reactive oxygen metabolites, thiol groups), inflammation (INFL; e.g., globulin, haptoglobin, ceruloplasmin and myeloperoxidase), and liver distress (e.g., gamma-glutamyl transferase, alkaline phosphatase, and paraoxonase) hematochemical parameters determined by the ILAB-60 auto-analyzer and, milk leucocyte subpopulations (immune system – IS) measured with flow cytometry (i.e., total milk leucocytes, polymorphonuclear cells, T-killer cells, T-helper cells, B cells and macrophages). These categories were submitted to the Data Integration Analysis for Biomarker discovery using Latent Components algorithm for the identification of highly correlated traits capable of discriminating the cows' infection status. The integration analysis highlighted a strong covariation between the OXI and INFL groups ($r^2 = 0.78$), and the MET and IS category ($r^2 = 0.68$). Among the most informative MET variables we found glucose, allantoin and acetate, the latter being negatively correlated with total leucocytes. Further analyses will help to identify hub variables able to discriminate sub-clinically infected animals. Acknowledgements: Funded by the European Union. Views and opinions expressed are however those of the author(s) only and do not necessarily reflect those of the European Union or REA. Neither the European Union nor REA can be held responsible for them.

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Theatre 3

Effects of feeding different microencapsulated sources of long and very-long chain unsaturated fatty acids on production and blood parameters in response to an immune challenge

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Omega-3 (n-3) fatty acids are known immunomodulators that attenuate inflammatory responses. Our goal was to evaluate the effects of feeding microencapsulated forms of fish, algae, and oleic acid oils on the response to an immune challenge. Nine Holstein cows (mean \pm SD: 82 \pm 18 DIM; 3.7 \pm 0.9 lactations) were randomly assigned to treatments in a replicated 3 \times 3 Latin Square design with 9-d periods and 7-d washouts. At d 9 of each period cows received an i.v. lipopolysaccharide (LPS) bolus (30 ng/kg of BW of *E. coli* O55:B5). Cows were milked twice daily. Blood was sampled at h 0, 2, 6, 12 and 24, relative to LPS challenge. Treatments were top-dressed daily and consisted of microencapsulated fish oil (FO; Prototype 6; 36% fish oil), algae oil (ALG; Prototype 39; 45% algae oil), and oleic acid (OA; Prototype 40; 20% oleic acid). The FO and ALG products delivered 25 g of the sum of n-3 (20:5 and 22:6), while OA provided 40 g of c9,18:1. Lipid supply was balanced at 226 g/cow/d. Data were analyzed under a mixed model with fixed effects of treatment, parity, time, and their interactions. Cows on FO and ALG tended to suffer a lower milk yield reduction than OA cows ($P=0.12$), and DMI was higher in FO relative to ALG ($P=0.06$). Milk fat content was higher in ALG relative to FO ($P<0.05$). No treatment effects were detected on plasma glucose, however, at h 2 ALG had lower levels of glucose than FO and OA cows ($P<0.05$). Plasma NEFA peaked at h 2 and 24 (Time, $P<0.05$) and TNF- α at h 6 (Time, $P<0.05$) with lower levels at h12 for ALG and FO compared to OA ($P<0.15$). Cows supplemented with ALG, tended to have lower insulin levels ($P=0.10$) and significantly higher Interleukin-1 β ($P<0.05$) compared to FO. Response to an immune challenge may depend upon chain length degree of saturation of the unsaturated fat supplements.

Alternative splicing mRNA isoforms linked to subclinical intramammary infection in Holstein cattle

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Alternative splicing could generate mRNA isoforms differentially expressed (DE) and play a key role in mastitis development. RNA-Sequencing using MGISEq platform from milk somatic cells samples from Holstein cows with no history of mastitis (n=9, NEG) and with natural subclinical intramammary infection (sIMI) due to *Streptococcus agalactiae* (n=11, Sa+) or *Prototheca* spp. (n=11, P+) was performed. DE mRNA isoforms (DEIs) among the groups using the *Bos Taurus* ARS-UCD1.3 reference genome were identified using CLC Genomics Workbench (FDR<0.05, |FC|>2). Functional analysis was performed using IPA (FDR<0.05). The NEGvsSa+ and NEGvsP+ comparisons revealed 16 and 27 annotated DEIs, 9 and 11 annotated DEIs with novel length and 1 and 7 novel DEIs with no previously annotated associated gene or length, respectively. Sa+vsP+ showed 5 annotated and 4 annotated with novel length DEIs. Functional analysis showed 54 significantly enriched metabolic pathways in NEGvsSa+. Among them, primary immunodeficiency signaling pathway (upregulated in Sa+) linked to increased susceptibility to infection was the most significant. The NEGvsP+ revealed 51 significantly enriched pathways including macrophage classical activation signaling (upregulated in P+), responsible to activate M1 macrophages typically linked to uncontrolled inflammation and tissue damage. Two annotated DEIs with novel length, BOLA-DOA_6 and GBP2_7 (both up-regulated in P+), play an important role in the macrophage classical activation signaling. Functional consequence analysis was performed to identify SNPs and INDELS leading to splice site events (SSEs). Three insertions located in KRT78 and CSN1S1 lead to SSEs responsible for alternative splicing of those DEIs. These results contribute to better understanding of defense mechanisms and immune responses that may provide new strategies to control sIMI in cattle. Acknowledgements: Agritech National Research Center funded by the European Union Next-Generation EU (Piano Nazionale di Ripresa e Resilienza (PNRR)–Missione 4, Componente 2, Investimento 1.4–D.D.1032 17/06/2022, CN00000022)

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Theatre 5

Preservation of milk in liquid nitrogen during collection does not affect RNA quality for RNA-seq analysis

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The present study describes the applicability of a method for milk sample preservation using liquid nitrogen during collection, to avoid immediate sample processing and to prevent RNA degradation, for its downstream RNA-seq analysis. The quality and integrity of the total RNA isolated from milk fat globules was assessed and analysed by independent equivalence tests to determine if the minimum requirements were achieved. The A260/280 and IQ values were in compliance with the reference values of 1.9–2.1 and ≥ 9 , respectively ($P < 0.05$). However, RIN values ranged between 2.1 to 5.7, and thus would not meet the generally accepted value of 7 ($P > 0.05$). This could be due to the large number of low molecular weight RNA fragments present in samples, which is inherent to sample type, rather than sample preservation in liquid nitrogen or RNA degradation. Likewise, the RNA concentration values were low (120.43 ± 86.26 ng/ μ L, 102.87 ± 58.53 and 109.43 ± 87.88 , measured by Nanodrop, HS Qubit and QuanTI Ribogreen, respectively). Despite that, after RNA-seq, an average of 52.7 million paired-end reads were obtained per sample. Although some overrepresented sequences were observed, the raw sequencing reads met the required quality control values for length (151 pb) and base-coverage (100%), base content (49%), and quality scores (average of 36), which would allow successful gene expression profiling. Thus, milk sample preservation using liquid nitrogen during collection does not appear to affect the quality and integrity of the RNA extracts isolated from milk fat globules and could be considered a more practical tool to improve the use of a non-invasive means of measuring the mammary epithelial transcriptome.

Hepatic mitochondrial function in dairy cows with different levels of pasture inclusion during lactation

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Mitochondrial function is crucial for the maintenance of energy homeostasis and a balanced stress response. To study the effect of pasture inclusion and stage of lactation on hepatic mitochondrial function a data set combining data from two published lactation studies including 68 cows fed with different percentages of grazed pasture inclusion in the diet (0, 30 or 100%; 0-PAS, 30-PAS or 100-PAS) evaluated at early and mid-late lactation (28 ± 10 vs. 215 ± 50 days in milk, STAGE) was used. Variables were grouped as: mitochondrial respiration parameters related and not related to ATP synthesis and energy balance markers and a principal factor analysis was performed with a total of 21 variables. Data were analyzed using a mixed model for the variables with the largest loadings (> 0.75), the model included, PAS, STAGE and the interaction PAS x STAGE. The first two main factors explained 57% of variation in the three analyses performed and the subset of variables obtained were: maximum respiration, coupling efficiency (COUP), state 4 respiration, leaking control ratio (LCR), beta-hydroxybutyrate (BHB) and energy corrected milk (ECM). The interaction PAS x STAGE ($P \leq 0.05$) was observed for COUP, LCR and ECM as COUP was the lowest for 0-PAS and 100-PAS during mid-late lactation, LCR the lowest for 0-PAS during early lactation and the highest for 100-PAS during mid-late lactation and ECM was the highest for 0-PAS during early lactation. Maximum and state 4 respiration were higher during mid-late lactation and BHB was higher during early lactation ($P < 0.05$) and higher for 100-PAS vs. 0-PAS ($P < 0.05$). Our results suggest that oxidative phosphorylation in hepatocytes is less efficient during mid-late lactation and for systems with a greater inclusion of pasture.

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Theatre 7

Oleic Acid Promotes Lipogenesis and Improves Mitochondrial Function in Bovine Adipocytes: The Role of PPAR α Activity

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In periparturient cows, oleic acid (OA) limits lipolysis and improves adipose tissue (AT) insulin sensitivity. In rodent models, OA enhances mitochondrial biogenesis and function. However, mechanisms behind OA effects in cows are unknown. Our objective was to determine the effect of OA on lipid accumulation and mitochondrial function and identify its mechanisms of action in bovine adipocytes. Pre-adipocytes were isolated from subcutaneous AT explants ($n=6$, non-lactating, non-gestating Holstein cows) and induced to differentiate into adipocytes in standard differentiation media (CON) supplemented with palmitic acid (PA), OA, or 60/40 mix of PA and OA (60-40) at $300 \mu\text{M}$ with or without PPAR α antagonist (GW6471; $10 \mu\text{M}$) for 7 d. PPAR α expression was determined by capillary electrophoresis (d 4). Triglyceride levels were quantified using Triglyceride-Glo Assay. Gene networks expression was evaluated using RT-qPCR. The statistical model included random effect of cow and fixed effect of treatment. OA and PA increased PPAR α protein expression compared with CON ($P < 0.02$). Compared with CON, supplementing fatty acids enhanced lipid accumulation ($P < 0.01$). However, PPAR α inhibition decreased OA and 60-40 triglyceride content ($P < 0.05$) but not for PA ($P = 0.26$). Within the mitochondria, PA tended to decrease expression of fatty acid transport protein system (CAC, CPT1, CPT2), complex I protein (NDUFS1), SIRT1, and PGC1 α compared with CON and OA ($P \leq 0.07$). Results show that OA restores mitochondrial biogenesis and improves oxidative phosphorylation. Our data provide mechanistic evidence for the use of OA in dairy cow diets during the periparturient period to enhance lipid accumulation. Improving energy balance will minimize health disorders and improve production of early postpartum cows.

The Stressogenic Effect of Bacteria Secretome is Modulated by the Size of the Milk Fat Globule Used as a Substrate

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Milk fat is secreted in milk fat globules (MFGs), a unique structure consisting of a triglyceride core covered with polar lipids and proteins derived from mammary epithelial cell (MEC) membranes. We previously demonstrated that mammary gland bacteria can thrive on these lipids and their growth and biofilm formation capabilities are dependent on MFG size used as a substrate, while releasing different chemical components (secretome) to the environment. This study aimed to investigate whether the response of MECs to the bacterial secretome is dependent on the MFG size used as a substrate for the bacteria. MFGs from raw milk were separated by size and used as a substrate for *Bacillus subtilis*, a commensal bacteria and *E. Coli*, a pathogenic bacteria. After 24 h, the secretome was collected, filtered, and used as a MEC conditioning media. The pro-inflammatory response and impact on lipid metabolism was recorded and compared with that of MECs treated with lipopolysaccharide (LPS) to assess stress levels. MECs exposed to large MFG-cultured *B. Subtilis* responded similarly to MECs exposed to LPS, which was reflected in increased pro-inflammatory gene expression and reduced lipid droplet diameter. MECs exposed to *E. coli* secretomes, showed an increase in pro-inflammatory gene expression regardless of the MFG size used as a substrate. These results indicate that MEC response to bacterial output is modulated by the size of MFGs that the bacteria can utilize in the mammary gland, which can impact the wellbeing and health of the cows as well as milk quality.

Immunometabolic responses of porcine blood immune cells to endotoxin and glucocorticoid stimulation

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In farm animals, little is known about the connection between energy metabolism of immune cells and their immunophenotype. To close this knowledge gap, we explored metabolism of porcine peripheral blood mononuclear cells (PBMC) challenged in vitro with pro- and anti-inflammatory stimulants, endotoxin (LPS) and dexamethasone (DEX), respectively. In the first experiment, we pretreated PBMC with increasing concentrations of a glycolysis inhibitor (2-deoxy-D glucose, 2-DG) or mitochondrial ATP synthesis inhibitor (oligomycin) for 30 min. Subsequently, the cells were treated either with vehicle, LPS, DEX, or LPS+DEX for 24h. The immune response was assessed based on the concentration of TNF- α , released by PBMC and modulating many inflammatory responses, using ELISA. While the inhibition of glycolysis by 2-DG significantly reduced the LPS-induced inflammatory response, oligomycin had no significant effect. In contrast, the anti-inflammatory effect of DEX is partially dependent on ATP synthesis but not on glycolysis, as oligomycin, but not 2-DG, showed dose-dependent effects. To study metabolic response to stimulation, in the second experiment we employed the Seahorse XFe96 flux analyzer to evaluate mitochondrial respiration and glycolysis, respectively, in PBMC challenged for 24h. Our results consistently demonstrated significantly higher glycolysis in LPS-treated PBMC compared to all other treatments, but provided no evidence for a change in mitochondrial respiration. In contrast, DEX significantly reduced LPS-induced glycolysis. Furthermore, DEX consistently reduced mitochondrial respiration and ATP production. Transcriptome analysis reveals the mechanisms how LPS and DEX modulate immunometabolism in PBMC.

Associations between social behavior and immune cell transcriptome in pregnant sows

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Social experience is an important predictor of disease susceptibility in social mammals. In the field of social genomics, a link between social stress, social dominance and immune cell gene expression have been demonstrated. However, the importance of non agonistic social behaviors, particularly positive interactions, remains unexplored. Thus, this study aimed to assess transcriptomic variations in peripheral blood mononuclear cells (PBMC) of pregnant sows exhibiting high or low levels of social exploratory behavior. Pregnant sows (n = 71) of mixed parities were raised in four stable groups of 17-19 animals during gestation (from gestation day (G) 0 to G105). The behavior was observed from G99 to G104, and animals were categorized based on their frequency of expression of nosing and grooming behaviors: High (HSE) or Low (LSE). Blood samples were collected at G98 on a subset of multiparous sows (HSE, n = 10; LSE, n = 11). PBMC transcriptome analysis revealed 102 differentially expressed genes (DEGs; adjusted p-value < 0.1, FC < 0.8 or > 1.2). Down-regulated DEGs in the HSE group were predominantly related to inflammatory pathways, such as inflammasome (CD36, NLRP3), TNF receptors superfamily (TNFRSF1A, TNFRSF1B, TNFSF13B), colony-stimulating factor (CSF2RA), MAP kinases (MAPK13, MAPK4, MAP3K20), and JAK-STAT (SOCS3). Whereas up-regulated DEGs encompassed functions like energy metabolism (ATP6V1C2, GRAMD1C, COQ8B), B cell activity (BLK, LIME1), nervous system (DLGAP4, ADRA2C), and others. In conclusion, our findings suggest that social behaviors are associated with transcriptomic changes in sows' PBMC, as indicated by alterations in the expression of genes particularly linked to inflammatory pathways.

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Theatre 11

Acetyl-CoA Carboxylase 1 Inhibition Reduces Fat Deposition Through a Histone Acetylation Signaling Axis in Pigs

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The thermogenic properties of beige adipocytes are crucial for combating human obesity and enhancing cold resistance in pigs. In our prior study, we exposed four local Chinese pig breeds to acute cold conditions (4 h at 4°C). We discovered that cold resistance in Tibetan and Min pigs, known for their robustness to cold, relies on the formation of beige adipocytes and the up-regulation of UCP3. In contrast, cold-sensitive breeds like Bama and Wuzhishan pigs lacked these cellular responses. Despite these findings, the molecular basis of beige adipocyte formation in pigs and its physiological consequences are still largely unexplored. Our recent study revealed that acute cold exposure inhibits the de novo lipogenesis (DNL) pathway in the adipose tissue of cold-resistant breeds, suggesting a potential role for this inhibition in beige adipocyte formation in these breeds. Furthermore, we observed that beige adipogenesis and DNL pathway down-regulation in cold-tolerant breeds might be depended on peripheral nerve activity. Notably, we identified that the thermogenic program in porcine adipocytes is governed by acetyl-CoA carboxylase 1 (ACACA), a rate-limiting enzyme in the DNL pathway. Manipulations of ACACA expression—either overexpression or knockdown—altered thermogenesis in porcine adipocytes, while treatment with the ACACA inhibitor 5-(tetradecyloxy)-2-furoic acid (TOFA) markedly boosted adipocyte thermogenesis. In vivo, TOFA treatment not only improved body temperature maintenance but also induced beige adipocyte formation during acute cold exposure in cold-sensitive weaned Bama pigs. Sustained TOFA administration for five weeks significantly enhanced carcass leanness (from 47.6 to 50%) and intramuscular fat content (from 1.21 to 1.54 g/100 mg), and reduced backfat thickness (from 1.74 to 1.53 cm) in 4-month-old Bama pigs. Mechanistically, ACACA inhibition promotes beige adipocyte formation by upregulating PRDM16 expression through enhanced acetylation of histone H3K9. These findings highlight the critical role of ACACA in modulating cold resistance and fat accumulation in pigs, presenting it as a significant molecular target for improving pig breed performance.

The hepatic gene expression of specific transporters of the BCAA and mTOR differs between North American and New Zealand Holstein cows under grazing conditions

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It has been reported that the better adaptive capacity to grazing conditions of New Zealand Holstein (NZH) compared with North American Holstein (NAH) cows was associated with lower labile protein mobilization and lower plasma concentrations of branched-chain amino acids (BCAA) during early lactation. High BCAA are associated with a chronic activation of the mTOR pathway and insulin resistance development in mammals. Despite BCAA bypass the liver, previous studies in mice demonstrated that the hepatic gene expression of some transporters of the LAT group, which are specific for BCAA, responded to nutritional status. We explored the effect of the Holstein genetic strain (GS) on the hepatic mRNA expression of LAT transporters, branched-chain fatty acid degradation enzymes and mTOR pathway. Gene expression of liver samples of NAH (n = 8) and NZH (n = 8) obtained at -45, 21, 100 and 180 days in milk (DIM) was determined by qPCR. Results were analysed as repeated measures and means differed if $p < 0.05$. The expression of LAT1 and LAT4 were affected by the interaction GS×DIM as it increased at 100 DIM only for NZH. The gene expression of mTOR, its effector S6KI, and the branched-chain fatty acids-degrading enzyme – BCKDH α – was greater for NAH than NZH during the experimental period, while LAT3 and BCKDH β mRNA were not affected by GS, DIM nor its interaction. Our results suggest that the BCAA could play signalling roles in the liver of dairy cows in relationship with its adaptive capacity to grazing conditions.

Prediction of ad libitum dry matter intake rebound after a feed restriction through cerebrospinal fluid amino acids concentration in dairy cows

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Dry matter intake (DMI) regulation is not yet fully understood in dairy cows. Some amino acids (AA) appear as potential compounds sensing the nutritional status and regulating the appetite in the central nervous system. We explored the prediction of DMI rebound after a feed restriction period through AA concentration on cerebrospinal fluid (CSF). Eight primiparous Holstein (18.5 ± 4.1 kg/d of milk yield, 472 ± 41 kg of BW, 2.5 ± 0.19 of BCS, 109 ± 28 DIM) housed in individual tie stalls (free available water) were fed a TMR diet formulated to meet requirements for maintenance and production during 14 days as follows: ad libitum offer (day 0 to 4), feed restriction to 50 % of the average DMI in the previous period (day 5 to 9), refeeding ad libitum (day 10 to 14). At day 9 the CSF samples were taken from the cerebellum-medullar cistern by puncture and AA concentrations were determined by gas chromatography time-of-flight mass spectrometry. Data was analyzed with Partial Least Squares Regression (PLS-R). The final PLSR model (1st component) showed acceptable predictive ability and goodness of fit ($Q^2=0.56$; $R^2Y=0.62$) and it comprised 11 AA as predictors of DMI (Cys, Pro, Phe, Asp, Tyr, Gly, His, Trp, Val, Asn, and Lys). Except for Cys that showed a positive association, the higher concentrations of the AA during the feed restriction period, the lower DMI rebound in the refeeding period. Our results suggest that these AA could play a signaling role in relation to energy generation, neuronal signaling and/or synthesis of neurotransmitters that affect central feed intake regulation.

Optimizing the bioenergetic parameters of peripheral blood mononuclear cells in dairy cows

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Mitochondria play vital roles in nutrient conversion to chemical energy and stress response mechanisms. Recent human research suggests that circulating peripheral blood mononuclear cells (PBMCs) may serve as indicators or biomarkers of mitochondrial function. Our objective was to establish an experimental protocol for PBMC isolation and mitochondrial function examination in dairy cows. Five Holstein dairy cows (193±35 days in milk, 561±87 kg live weight, 25.4±5.9 kg/d average milk yield) were used, and 8 mL of blood was drawn from the coccygeal vein to isolate PBMCs using the gradient method. Mitochondrial oxygen consumption rates were measured using 2 million PBMCs/mL in the Oroboros Oxygraph-2k with oligomycin (ATP synthase inhibitor), FCCP (oxidative phosphorylation uncoupler), and antimycin and rotenone (respiratory chain inhibitors), and the respiratory control ratio (RCR) was determined. Plasma glucose, urea, cholesterol, insulin, and glucagon were quantified using commercial kits. Principal factor analysis was performed using the PROC FACTOR procedure in SAS. The RCR was 8.8±3.0, glucose 4.1±0.1, urea 4.1±0.3, cholesterol 6.9±0.4 mmol/L, and the insulin/glucagon ratio was 2.4±0.6. The principal factor analysis showed that 75% of the variation was explained by the first two factors, and the variables with the highest loadings were the insulin/glucagon ratio (0.95) and RCR (0.97), with a significant and positive correlation found between RCR and plasma glucagon ($r=0.9$ and $P<0.05$). In conclusion, our preliminary results highlight the significant correlation between PBMC mitochondrial function and the insulin/glucagon ratio of dairy cows, demonstrating the potential of these cells as biomarkers. However, further studies are needed to fully elucidate the mechanisms of these cell biomarker in dairy cows

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Diet-induced changes in the jejunal microbiota of broilers during development, greatly affecting *Enterococcus hirae* and *Enterococcus faecium*

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Modern broiler breeds allow for high feed efficiency and rapid growth, but less attention is given to resilience against pathogens and disease. Broiler growth rate, feed efficiency, and health are affected by the composition of the gut microbiota, which in turn is influenced by diet. In this study we therefore assessed how diet composition can affect the broiler jejunal gut microbiota. A total of 96 broiler chickens were divided into four diet groups: control, coated butyrate supplementation, medium-chain fatty acid supplementation, or a high-fibre low-protein content. Diet groups were sub-divided into age groups (4, 12 and 33 days of age) resulting in groups of 8 broilers per diet per age. The jejunum content was used for metagenomic shotgun sequencing to determine the microbiota composition on species level. The composed diets resulted in a total of 104 differentially abundant bacterial species. Most notably were the butyrate-induced changes in the jejunal microbiota of broilers 4 days post-hatch, resulting in the reduced relative abundance of mainly *Enterococcus faecium* and the opportunistic pathogen *Enterococcus hirae*, when compared to the control diet. This effect takes place during early broiler development, which is critical for broiler health, thus exemplifying the importance of how diet can influence the microbiota composition and its relation to broiler health. Future studies should therefore elucidate how diet can be used to promote a beneficial microbiota in the early stages of broiler development.

Studying The Impact Of A Standardized Dry Grape Extract On Pullets' Metabolome

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Vitis vinifera extracts are powerful antioxidants widely used to treat oxidative stress. Several studies have described the positive impact of a standardized dry grape extract (SDGE) as feed additive on the antioxidant status in poultry. However, research into the mode of action of SDGE to guarantee this effect is scarce. In this study, a non-targeted metabolomic approach was used to investigate the effect of a dietary supplementation of SDGE on pullets' metabolic pathways. 36300-day-old pullets were randomly divided in two barns on the same farm site. Both received the same diet, with the addition of 30ppm of Nor-Grape[®] (Nor-Feed, France) in the feed of the supplemented group (NG, n=18150) from D1 until W17. On W17, blood samples were drawn from 8 pullets per group. Metabolites extraction was carried out using two-step extraction protocol. Two extracts from each sample (polar and apolar extracts) were analyzed. Progenesis QI was used for data processing, identification and statistical analysis (principal component analysis (PCA)). IMPaLA was used for metabolic pathways analysis. PCA results revealed distinct profiles between groups, with 574 differently expressed features ($p < 0,05$, max fold change > 2). Among them, 70 were identified with an acceptable score. Metabolic pathways analysis evidenced a global impact of the SDGE supplementation on cell signaling with specific effects pathways such as glutamyl cycle and GPCR. Thus, dietary supplementation of 30ppm SDGE modulates pullets' plasma metabolome. The metabolic pathway analysis suggests an implication of SDGE in the modulation of several pathways implicated in the antioxidant system as well as other aspects, such as GPCR.

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Antibiotic resistome of commercial Lohman classic layers with and without mannan rich fraction supplementation

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This work aimed to identify antibiotic resistance genes (ARGs) present in the cecum of commercial layers and to understand the impact of dietary supplementation on the resistome. ARGs move between bacteria in the gut of animals and to bacteria that may cause infections in animals or humans. Limiting the presence and abundance of ARGs within the intestine may help limit ARG transfer among bacteria. DNA was extracted from cecal content of 12 birds per shed (77 weeks old) fed with either a commercial diet (CON) or CON plus MRF (1 kg t⁻¹) (Alltech Inc). Sequencing libraries were sequenced on an Illumina NovaSeq 6000 platform with paired end 150bp reads. The Resistance Gene Identifier software was used to align Unigenes with the Comprehensive Antibiotic Resistance Database to analyse the core (CR) and accessory resistomes (AR). Metastats analysis was used to perform permutation tests between groups. The p-value was corrected using the Benjamini-Hochberg False Discovery Rate. 468 ARGs were identified between both dietary groups and each group had 450 identified ARGs. For CON, 68 ARGs were assigned to the CR and 382 ARGs were assigned to the AR. In MRF, 63 ARGs were assigned to the CR and 387 ARGs were assigned to the AR. Within the CR, 30 genes were unique to CON and 25 genes unique to MRF. Within the AR 43 genes were unique to CON and 48 genes unique to MRF. Tetracycline resistance genes were most abundant in both groups. A total of 49 ARGs were shown to be different between groups ($P < 0.05$, $q < 0.05$). This study provided a survey of the CR and AR of commercial layer cecal content, showing MRF supplementation influenced resistome composition and relative abundance. These data may be used to further the development of feed supplements to lower the presence of ARGs within poultry.

Boosting underperforming broilers: feed structure strategies for better gut development and health, and microbiotaM. Z. Akram¹, E. A. Sureda¹, N. Everaert¹¹ KU Leuven, Department of Biosystems, 3001 Leuven, Belgium

Despite fine-tuned genetics and nutrition, body weight (BW) heterogeneity persists within broiler flocks. Underperforming chicks often face delayed gut development and sub-optimal microbial colonization, hindering them from reaching their genetic potential. This study investigates the effect of in-feed structural components, such as coarse corn (CC), oat hulls (OH), and their combination (CO), to enhance the performance of underperforming broilers. On day (d) 7, 1400 male broiler chicks, were categorized based on BW. Low BW chicks (n=504 chicks) were assigned to four dietary groups: standard diet (LBWC), diet with 7% CC (LBW+CC), diet with 3% OH (LBW+OH) and combination of both (LBW+CO). High BW chicks (n=126) exclusively received the standard diet (HBWC). Diets were isoenergetic and isonitrogenous and offered in two phases: Grower (d 8 to 16) and Finisher (d 17 to 38). Data on performance, digestive organs, and gut permeability were collected on d 14, 21, and 38, analyzed by one-way ANOVA. Caecal microbiota was analyzed on d 38 by 16s rRNA gene sequencing. HBWC group consistently outperformed the LBWC group. By d 38, LBW birds on dietary groups had numerically higher BW than the LBWC group with no statistically significant difference from HBWC, indicating the potential of physical structure in feeds. Gizzard size on d 21 and 38 was increased in LBW +CC, +OH, and +CO groups compared to LBWC and HBWC groups. LBWC had significantly higher gut permeability than HBWC, while LBW groups with dietary treatments showed a numerical decrease in gut permeability. *Lactobacillus* was enriched in LBW+CC, *Romboutsia* in LBW+CO, and *Fournierella* in LBW+OH groups. Thus, adding structural components to diets improved the performance of LBW chicks by influencing gizzard development, gut permeability, and caecal microbiota.

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Effect of *Urtica dioica* and *Urtica urens* on survival in broilers chickensJ. Teixeira^{1,2,3}, Â. Martins^{1,2,3}, J. L. Mourão^{1,2,3}, A. Alves^{1,2,3}, V. Pinheiro^{1,2,3}¹ UTAD, Qta. Prados, 5001-801 Vila Real, Portugal, ² CECAV, Qta. Prados, 5001-501 Vila Real, Portugal, ³ AI4Animals, Qta. Prados, 5001-501 Vila Real, Portugal

The utilization of nettles in animal feeding is deeply linked to their traditional use as preventatives for disorders in chickens. In Portugal, the most common nettle species are *Urtica dioica* and *Urtica urens*, which have been associated with alternatives to mitigate the incidence of digestive pathologies in broiler chickens by improving intestinal health, potentially increasing flock survival. Further research is necessary on such alternatives and efficient products to enhance intestinal microbiota and understand their role in the immune system. In poultry farming, bird mortality represents high production costs, thus it is crucial to implement strategies that contribute to reducing this productivity parameter. The aim of this study was to investigate the effect of two nettle species on the survival of meat chickens and diagnose the causes of mortality through post-mortem necropsy. 240 male chicks of the Ross 308 AP strain, aged zero days, were used and monitored until 24 days of age. They were housed in 30 pens with 8 birds each, and divided into 3 treatments (10 replicates per treatment): Control basal diet (CT); basal diet with 1% incorporation of *Urtica dioica* (UD); basal diet with 1% incorporation of *Urtica urens* (UU). Over the 24-day trial period, 219 birds survived, and significant differences were observed (P=0.0174 Fisher Exact Test) when comparing the CT group with the UU group, with Odds ratio of 6.22 and confidence interval between 1.33 and 29.03, indicating that birds in the CT group had a mortality rate 6 times higher than the UU treatment. No significant differences were observed when comparing the CT group with the UD group. Of the 21 dead birds, Ascites was the cause of death in 19 cases. It can be concluded that *Urtica urens* has a significant effect on the survival of broiler chickens, while *Urtica dioica* did not show any difference. Acknowledgments: This work was supported by the projects UIDB/00772/2020 (Doi:10.54499/UIDB/00772/2020) funded by the Portuguese Foundation for Science and Technology (FCT).

Nurturing Progeny: Probiotics' Influence on Sow Gestation Performance and Piglet Weaning Weight

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Probiotics benefit sow immune systems, important to minimize reproductive losses during farrowing in pig production. We aimed to evaluate the effects of probiotic on sows' gestation performance and piglet's weaning weight. A total of 47 sows from crossed commercial lines were randomly allocated in four treatments according to their farrowing order (from 1 to 6 farrowings or from 7 to 12 farrowings). Treatments were: T1) control group – basal diet without probiotic (n=11); T2) basal diet + addition of probiotic throughout the entire gestational (1st day) and lactational period (n=12); T3) basal diet + addition of probiotic from the second third of gestation (38th day) and lactational period (n=12); and T4) basal diet + addition of probiotic from the last third of gestation (76th day) and lactational period (n=12). The probiotic (*Enterococcus faecium*, *Lactobacillus acidophilus*, and *Lactobacillus plantarum*) was administered daily according to the reproduction phase, with gestation receiving 1g/sow/day and lactation receiving 6g/sow/day. The following parameters were recorded: total number of born, live born, stillborn, crushed piglets, and piglet weight at birth. For all the piglets born, half received, orally, 2mL with 0.5g of a new probiotic (*Bifidobacterium bifidum* and *Lactobacillus casei*), considered the probiotic treatment group, and the other half received water in equal measure as a placebo, being considered the control treatment group. Later, their weaning weight was recorded. All data was analyzed through ANOVA, with means evaluated by the Tukey test. No statistical difference was observed among treatments for sows' gestation performance ($P>0.05$). Sows from T4 had heavier piglets in weaning compared to all other treatments ($P<0.01$), independent of piglet's treatment. Piglets that received probiotics were majorly heavier than those that did not receive them ($P<0.01$), independent of sows' treatment. Data showed that probiotics can improve piglet weaning weight, with the most significant impact observed through direct administration or when administered during the last third of the sows' gestation.

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Effect of replacement of soybean by Mediterranean legumes on intestinal health of growing pigs

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The effect of soybean protein replacement by local legumes in diets for growing pigs was evaluated for intestinal health. Calprotectin, an indicator of inflammation, and cecal volatile fatty acids (VFA) were analyzed. Local peas, lentils and chickpeas replaced 50 or 100% of dietary protein from soybean (pea100, pea50, lentil100, lentil50, chickpea100, chickpea50) and they were compared with a soybean control. A total of 252 pigs (20.3 ± 1.34 kg) were allocated in 7 experimental treatments (6 replicates/treatment; 6 pigs/replicate). Diets were isonutritive (13.1 MJ/kg ME and 16.2% CP). Fecal samples (1 sample/replicate) were obtained on days 0, 14 and 28 to determine calprotectin. Cecum content was sampled on day 28 (4 samples/treatment) to determine VFA. For statistical analysis, contrasts were conducted between control and each legume. Calprotectin values were not different from control for any legume ($P>0.05$) at any time. However, chickpeas increased total VFA, acetate, propionate, butyrate, and decreased ($P<0.05$) the sum of isoacids (isobutyrate and isovalerate, plus valerate) and branched chain ratio (ratio of isoacids to straight-chain VFA; BCR). BCR indicates the fraction of VFA, related to protein fermentation. Peas tended to increase total VFA, acetate and propionate, tended to decrease BCR and decreased isobutyrate and isovalerate. Lentil tended to increase butyrate and decreased BCR, isobutyrate and isovalerate ($P<0.05$). Increased butyrate and straight chain VFA and decreased BCR indicated enhanced intestinal health of local legumes compared to soybean. Funded by European Union's Horizon Europe Research and Innovation Program, Grant agreement No. 01059609 (Re-Livestock).

Effect of dietary supplementation of *A. nodosum* and *L. calcareum* combination on piglets' health

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Livestock will have to face challenges aimed at sustainable use of land and resources, ensuring the maintenance of the production system, and reducing antimicrobial resistance. Therefore, in line with One Health principles, functional nutrition could promote animal health and decrease the risk of pathology development. The following study aimed to assess the effect of an innovative dietary supplementation of the combination of *Ascophyllum nodosum* and *Lithothamnium calcareum* on piglets' health. Twenty-fourth post-weaning pigs (28±2 days) were enrolled in two different groups (n=12/group) balanced per weight and sex: control group (CTRL) was fed a commercial diet, and treatment group (ALGAE) was fed commercial diet supplemented with 1.5% of *A. nodosum* and 0.5% of *L. calcareum* for 27 days. Weekly, zootechnical performances were assessed by monitoring the body weight and feed intake. Fecal samples were collected weekly to evaluate the abundance of total, lactic acid and coliform bacteria through plate counting with selective media. Serum samples were obtained at day 0 and day 27 to assess the antioxidant barrier using colorimetric assays. Body weight showed no significant differences throughout the experimental period. A comparable trend for total, coliform, and lactic acid bacteria CFU in fecal samples of each group was observed over 27 days. A significantly higher serum antioxidant barrier was registered in ALGAE compared to CTRL group at day 27 (363.26 ± 16.24 vs 230.69 ± 32.08 HClO/mL, p < 0.05). In conclusion, the supplementation *A. nodosum* and *L. calcareum* could be considered a promising dietary strategy to enhance the oxidative barrier in weaned piglets.

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Use of apple pomace in pigs diet

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Progressing inflation and the increase in prices of animal feed give breeders an incentive to look for various ways to reduce production costs. Poland is the largest apple producer in Europe, and approximately 20-40% of the total apple production is processed. By-products include, among others: pomace, peel and seeds, which constitute from 10 to 35% of the weight of the raw material, and their disposal constitutes a significant burden for the food industry. Numerous studies indicate the possibility of using fruit pomace in various industries, including the feed industry, which is consistent with the idea of sustainable agriculture. Due to its chemical composition, apple pomace may be a valuable source of fiber, minerals, vitamins, lipids, antioxidants and pectins, and their use in feed for fattening pigs may have a beneficial effect on animal performance, the development of the digestive tract and the quality of pork. The aim of the study was to assess the effects of introducing dried apple pomace into the diet on the production parameters of fattening pigs and on post-slaughter indicators of carcasses. Based on preliminary research, it was noticed that the addition of apple pomace to feed for fattening pigs reduces the growth of pigs and their slaughter weight. There was no deterioration in the feed conversion rate. The addition of apple pomace in pigs diets will have a positive effect on the post-slaughter parameters of the carcass and the slaughter efficiency. Research financed by the EU – Horizon 2020 "mEATquality" project no. 101000344.

Butyric acid with resistant potato starch improved growth and health of weaned pigs

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Butyric acid (BA) is the main energy source for colonocytes. It can be produced endogenously by microbial fermentation, and exogenous forms can also be supplemented in diets. Resistant potato starch (PS) is an additive that can be used by colonic microbes to produce short-chain fatty acids such as BA. This study aimed to compare the effects of supplementing exogenous BA versus promoting endogenous BA production, on the health and growth of weaned pigs. The 96 pigs were sorted into 4 groups and fed for 35 days: (1) control diet (C); (2) C+1.5% BA; (3) C+1% PS; (4) C+1.5% BA+1% PS (BAPS). A faecal score (FS) system of 1-5 was used to monitor diarrhoea (1=very hard, 5=very loose). There was a BA x PS interaction ($P<0.05$) on daily gain (DG), daily feed intake (FI), final body weight (FBW) and FS. The addition of BA had no effect on DG (510 vs 536, SEM 0.025) or FBW (24.4 vs 25.4, SEM 0.627) compared to the C diet. However, combining BA and PS increased DG (585 vs 518, SEM 0.025) and FBW (27.1 vs 24.9, SEM 0.627) compared to the PS diet. The addition of PS reduced FI compared to the C diet (773 vs 853, SEM 0.027), but combining PS and BA increased FI compared to the BA diet (922 vs 812, SEM 0.027). The addition of BA reduced FS compared to the C diet (2.35 vs 2.50, SEM 0.031), but combining BA and PS increased FS compared to the PS diet (2.27 vs 2.17, SEM 0.031). The inclusion of PS improved ($P=0.05$) FCR (1.53 vs 1.79, SEM 0.101) compared to non-PS supplemented diets. In conclusion, the combination of potato starch and butyric acid improved daily gain, feed intake and final body weight compared to the control and the individual supplementation of BA and PS. While the inclusion of potato starch improved feed conversion ratio.

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Intestinal barrier function of piglets fed AGP free diets supplemented with an association of fatty acids, prebiotics and zinc proteinate

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To evaluate the association of fatty acids, prebiotics and zinc proteinate (FPZ, Viligen™, Alltech) in piglet diets without AGP, on tight junction gene expression and microorganism translocation to the liver, eighty 21-d old piglets (6.69 ± 0.19 kg) were distributed in a randomized blocks design, with five treatments and eight replicates (a male and a female). Treatments consisted of a positive control (PC) diet (0.02% Halquinol), diet without AGP or FPZ, and three diets containing 0.10, 0.14 or 0.18% FPZ. Diets were phase fed to piglets from 21 to 35-d and 36 to 49-d old. The jejunum was collected from 49-d old piglets to evaluate the gene expression (qPCR) of zonula occludens (ZO), occludin (OCLN) and claudins (CLDN). Liver samples (49-d old) were collected, diluted and aerobically cultured on plates with MRS and VRB agar, for subsequent plate counting (CFU) to evaluate the translocation of *Lactobacillus* spp and *Enterobacteria* spp. The PC and 0.10 or 0.14% FPZ had the same CLDN3 ($P=0.0008$) and ZO1 ($P=0.0205$) expressions. The PC showed higher gene expressions for CLDN2 ($P=0.0001$) and CLDN5 ($P=0.0001$), while 0.14 or 0.18% FPZ resulted in lower expression of ZO2 ($P=0.0001$) compared with PC. Linear response plateau predicted lower ZO2 expression above 0.098% FPZ and the quadratic model estimated the highest expression of CLDN2, CLDN3 and CLDN5 at 0.081; 0.107 and 0.08% FPZ. No differences ($P>0.05$) were observed regarding the CFU of *Lactobacillus* spp and *Enterobacteria* spp in the liver. Therefore, under the conditions of this experiment, supplementing FPZ in piglet diets without AGP was effective to maintain the function of intestinal barrier.

Dietary inclusion of fibrous corn silages reduces the severity of gastric mucosa damage in heavy pigs
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The aim was to examine the impact of dietary inclusion of corn silages on productive performance of Italian heavy pigs and to evaluate a possible mitigation effect on gastric mucosa damages. A standard corn-soya pig diet was compared with a diet containing whole ear corn silage and high chopped whole plant corn silage (20 and 10% dry matter, respectively). In two consecutive feeding phases of 72 day each, 36 "Large White x Landrace" pigs (18 each phase) were divided into 6 homogeneous groups of three pigs with initial body weight (128 ± 15 kg), and kept in partially-slatted pens till slaughtering (final weight 183 ± 12 kg). Animal performance, meat quality, and stomach characteristics were analyzed by a completely randomized 2×2 factorial design with the diet as a fixed effect. Overall, the growth performances were high (on average 764 g/d), although the diet with corn silages determined a reduction trend ($p = 0.08$) in the growth rate of about 5-6% accompanied by a decrement in the organic matter digestibility (85.23 vs 81.93, $p < 0.01$). These aspects did not compromise the slaughter traits and the back fat fatty acids profile. Stomach characteristics were affected by diet containing corn silages, with an increase of stomach empty weight (1222 vs 1106 g, $p < 0.01$) and the oesophageal area (OA) surface (5.63 vs 4.60 %, $p < 0.01$). Moreover, silage dietary inclusion reduced the severity of stomach damages in the OA ($p < 0.01$). In conclusion, corn silages diets sustain heavy pig performance while reducing the severity of gastric mucosa injury. (Financed by Consorzio del Prosciutto and by IFCQ Certificazioni srl di San Daniele, Italy; supported by Pioneer, Cremona, Italy).

Algal-based extracts and their mixes: ability to perform antimicrobial and antioxidant activities

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Reducing the use of antibiotics in livestock to decrease the spread of antimicrobial resistance is today's challenge in modern farming system. Seaweeds are source of bio-active compounds for application as functional feed ingredients due to their antimicrobial and antioxidant activity. Thus, the aim of the study were to test the bio-activities of *Ascophyllum nodosum* (AN), *Palmaria palmata* (PP) and their 1:1 mixes (ANPP) via: (i) investigating the antimicrobial activity against enterotoxigenic *Escherichia coli* (ETEC) F4+ and F18+ and (ii) testing their antioxidant activity through chemical-based assays. Briefly, a culture-based ETEC assay was performed to evaluate the inhibitory activity with the treatments: 2 (with or without ETEC) \times 2 (F4+ and F18+) \times 5 doses (0, 1.44, 2.87, 5.75, 11.50, 23.0 mg/mL). Overnight-grown of ETEC F4+ and F18+ were treated with AN, PP, ANPP, measured (OD600, 60 min intervals) and quantified (CFU/mL). Besides, the antioxidant activity of different doses of AN, PP, ANPP (0, 1, 50, 100, 200, 500, 600 mg/mL) was assessed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging and reducing power (RPA) assays. All data were analyzed (SAS 9.4). AN and PP species dose-dependently significantly inhibited ($p < 0.05$) the growth of ETEC F4+ and F18+ in vitro. ANPP did not inhibit the ETEC growth. The results also exhibited antioxidant activity in DPPH and RPA assays, with AN as the strongest dose-response manner. Thus, tested algal species may be candidate as in-feed additives to antibiotics overuse in rearing system.

Development of nanocarrier for engineered *L. lactis* delivery as edible vaccine against swine *E. coli*

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Lactococcus lactis has been promoted as interesting platform for antigens expression. However, concerns persist about the resistance of antigens during the gastrointestinal transit. Halloysite nanotubes (HNTs) is a green nanomaterial that could be used for the protection and design of nanocarrier agents for targeted delivery. The aim of this study was to develop a nanocarrier platform for mucosal delivery of based on engineered *L. lactis* as model for multiepitope edible vaccines against major swine *E. coli* pathotypes. Specifically, *L. lactis* strains were engineered for the single expression of F4 adhesive fimbriae (FaeG), F18 adhesive fimbriae (FedF), and the B subunit of verocytotoxin 2e (Vte2-B) as immunogenic antigens for a probiotic-based vaccinal platform. Our approach involved designing vectors for the protein expression both intracellularly and anchored to the cell wall and protecting the obtained products with patent (n° 102021000006506). HNTs have been functionalized via a solid/liquid adsorption process in aqueous solution to preserve antigenic integrity and targeted release in the gut. The uptake of *L. lactis* was studied in the range of 0.20-1.5 mg/mL. Preliminary results showed an uptake of 15.7 ± 0.07 mgL. *lactis*/gHNTs, compared to the initial 33.75 mgL. *lactis*/gHNTs, achieving a reaction yield of 46.5%. This innovation constitutes a virtuous platform that combines nanomaterials and biotechnologies for reducing antibiotic use. The developed model represents a novel nanocarrier agents for oral vaccination in line with One Health principles.

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Poster 15

Relationship between CF or NDF in feed and content of fatty acids in sow feces

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The analysis of structural carbohydrates via NDF and ADF has been emphasized in the analysis of pig feed in order to better meet the requirements for intestinal health and thus also animal welfare. The nutritional situation of sows appears to be very complex. It is therefore necessary to check whether fecal sample analyses are suitable for describing the fermentation pattern. 5 companies were selected for the investigations. The rations of sows in the transit phase were then analyzed and the faecal samples of at least 6 sows per farm were also recorded. The feed and fecal samples were analyzed in an external laboratory. In addition to fatty acids, the levels of nitrogen and selected minerals were also analyzed in the faeces. On average across the farms, the rations in the transit phase had a mean value of 60 g CF and 201 g NDF per kg DM, with variation between farms ranging from 162 g to 239 g NDF. There were no differences between the farms in terms of manure particle distribution, but more than 80% of the manure had a particle size of less than 0.063 mm. The ratio of the fatty acids acetic, propionic and butyric acid in the feces of the sows was on average 60 % – 24 % – 16 %. However, strong variations in the SCFA content of the feces ranging from 2.1 g to 7.0 g per kg DM could be detected. Correlations were found between the levels of fatty acids in the feces and the NDF content of the feed (NDF and acetic fatty: $r = + 0.839$; $p = 0.005$). An exponential regression with a coefficient of determination of 79 % was calculated between the NDF content of the feed and the concentration of acetic acid in feces. In conclusion, it can be said that the levels of volatile fatty acids in the feces can be used to derive a certain statement about the effect of the NDF content in the feed.

Improving lactational performance in primiparous sows through targeted dietary supplementation for liver health

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From late gestation until weaning, the sow's liver is challenged, due to increased oxidative stress and nutrient processing requirements, especially those related to one-carbon metabolism. This may be more pronounced in primiparous sows. We assessed the effects of supplementing sows from day 84 of gestation to weaning with a hepatoprotective solution containing plant extracts, possessing liver antioxidant and anti-inflammatory activities, alongside B-complex vitamins and betaine. We hypothesized that supplementation would improve gestational and lactational performance along with liver health. Twelve primiparous sows were divided to control and supplemented (S) groups. Sow BW and backfat thickness (BF), were measured at days 84 and 109 of gestation and at weaning (day 21). Blood samples collected on day 4 post-farrowing were analysed for liver health indicators (Haptoglobin, ALP, AST, ALT, CK, and GGT). Reproductive performance metrics, such as number of piglets born alive, mummified, litter weight, and FI over gestation and lactation were recorded. There was no effect of treatment on sow BF, BW and FI during gestation and on the total number of piglets born, but S sows tended to have less mummified piglets ($P=0.08$). Amongst analysed markers Haptoglobin and ALT were significantly reduced (-42% and -13%, respectively; $P<0.05$) and ALP tended to be reduced in S sows (-18%; $P=0.07$). During lactation, supplementation significantly increased sow FI (+29%; $P=0.01$), reduced the loss in BF in lactation, and increased piglet ADG (+17%; $P<0.05$). In conclusion, this nutritional strategy improved liver health in primiparous sows, leading primarily to enhanced lactational performance.

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Poster 17

The impact of glutathione precursors on growth performance, nutrient digestibility, and blood profile in weaning pigs

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Glutathione (GSH) is an endogenous antioxidant found in all eukaryotic cells and plays an important role in maintaining redox homeostasis. Higher GSH concentrations protect against cellular damage, tissue degeneration, and disease progression in various animals, however, very limited studies were presented in pig models. Thus, we aimed to examine the impact of glutathione precursors on productivity, digestive performance, and blood profile in crossbred weaning pigs. Based on initial body weight 7.94 ± 1.53 kg and sex ((2 barrows and 3 gilts) $n=180$ pigs were randomly assigned to one of four dietary treatments for 42 days. The test treatments were: CON- basal diet, and the basal diet supplemented with 0.10%, 0.20%, and 0.30% of GSH. Each treatment has 9 replicates. Experimental data were analyzed using GLM procedure of SAS and the linear and quadratic test was used to determine the significance between treatment means ($P<0.05$). The inclusion of GSH precursor had linearly increased ($P<0.05$) the body weight of pigs on days 7, 21, and 42. Compared to CON, GSH supplemented group showed increased ($p < 0.05$) average daily gain (ADG) and gain to feed ratio (G/F) at d 42 and the overall experimental period. However, there were no improvements observed in nutrient digestibility and blood profile in the treatment group. From this, we infer that adding graded level GSH in pigs' diet would be more beneficial to enhance their growth rate without showing any adverse effect on other parameters.

Carry-on effect of fenugreek cotyledons on sows to their piglets during lactation

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Sow's nutrition is known to have important effects on their newborns' performances during their early life. This present study was aimed to assess the impacts of a commercial feed additive based on fenugreek cotyledons (Norponin® Cotyl, Nor-Feed, France) supplemented in late gestation and during lactation diets, on performance of sows and their piglets. Briefly, 12 multiparous sows (Landrace X Yorkshire) were randomly divided into 2 groups: control group (CT) fed with a standard gestation and lactation diets; supplemented group (COTYL) received the same diets supplemented fenugreek cotyledons at 2000 ppm from 9 days pre-farrowing continuing until weaning (21 days). Sows' average daily feed intake (ADFI expressed in kg per sow) was recorded. Sows' feces were sampled twice, at farrowing and at weaning, to measure apparent total tract digestibility (ATTD expressed in percentage). Average piglets body weight (kg) was compared at birth and weaning. No difference in ADFI was observed between groups, either in late gestation (1.93 vs 1.93, COTYL vs CTL respectively) or lactation (6.06 vs 6.32 respectively). Similarly, ATTD between 2 groups observed at farrowing was identical. However, at the end of lactation COTYL sows showed significantly higher ($P < 0.05$) ATTD of nitrogen (73.66 vs 68.85) and energy (73.59 vs 68.75) compared to CT sows. Numerical difference in piglets' birthweight was not significant (1.48 kg in CT group vs 1.68kg in COTYL group). In contrast, piglets from COTYL sows were significantly heavier ($P < 0.05$) at weaning (6.72kg) in comparison to the others (5.99kg). The results obtained from this trial show that the COTYL supplementation contributes to significantly increased feed digestibility in sow resulting in improved piglet's growth during lactation. Further studies are necessary to confirm these results.

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Poster 19

Effect of dietary *Bacillus subtilis* and *Bacillus licheniformis* probiotic on growth performance, nutrient digestibility and fecal microbiota in weaning pigs

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Probiotics have been used to address antibiotic alternatives and environmental concerns. Thus, the focal aim of this study is to evaluate the effects of probiotic complex supplementation on growth performance, nutrient digestibility, and faecal microflora in weaning pigs. A total of 120 [(Yorkshire × Landrace) × Duroc, 28 days old] pigs were randomly allotted to one of four dietary treatments for 6 weeks. The test treatments consisted of basal diet (CON) and basal diet supplemented with 0.1 %, 0.2 %, and 0.3 % of probiotic complex (Powerzyme100) consisting of *Bacillus subtilis* (1.0×10^9 cfu/g) and *Bacillus licheniformis* (1.0×10^9 cfu/g). Experimental data were analyzed using GLM procedure of SAS and the Linear and quadratic test was used to determine the significance between treatment means ($P < 0.05$). During the overall experimental period, pigs fed a diet supplemented with graded levels of the probiotic complex showed linearly increased ($P < 0.05$) average daily gain (ADG) and gain: feed ratio (G: F) ratio. Moreover, a linear increase ($P < 0.05$) in digestibility of dry matter (DM), nitrogen (N), and energy was observed in pigs fed probiotic complex. Compared to CON, pigs fed a diet supplemented with probiotic complex showed increased ($P < 0.05$) *Lactobacillus* and decreased *E. coli* counts. Our results demonstrate that adding probiotic complex supplements would serve as potential alternatives to AGP thereby improving growth performance, nutrient digestibility and reducing *E. coli* counts in weanlings.

The efficacy of the emulsifier DiNAMIC in growing pig diets

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DiNAMIC is a specific mix of lysophospholipids. In the gastro-intestinal tract DiNAMIC behaves as a classic bio-surfactant – breaking down the fats into more digestible emulsions. The size of the DiNAMIC molecule enables very small liposomes to form and these can carry a payload of fat and other nutrients across the gut villi membranes. This study investigated the effects of the addition of DiNAMIC in the grower ration offered to pigs from 7-10 weeks of age where oil inclusion is lower than in creep and link rations offered from 4-7 weeks of age. Eighty-four pigs were allocated at weaning into groups of six balanced for weight, gender and sire. Treatments: Control – standard creep (1.62% Lys, 15.8DE MJ/kg), link (1.50% Lys, 15.4DE MJ/kg), and grower (1.0% Lys, 14.5DE MJ/kg). Treatment –standard creep and link and grower + 750g DiNAMIC. Pigs were weighed at weaning (4 weeks of age), 7 weeks and 10 weeks. No significant differences were observed in the 4–7-week period as expected. DiNAMIC inclusion in the grower ration offered from 7-10 weeks of age had no effect on feed intake but significantly improved weight (32.5kg vs 30.8) growth rate (771g/d vs 705g/d) and FCR (1.37 vs 1.52) as hypothesised. This demonstrates that DiNAMIC inclusion assists in the digestion and absorption of nutrients, driving growth rate and feed efficiency when included in the grower ration.

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Poster 21

Phytase supplementation on weaned piglet diets: impacts on short-chain fatty acid production and gastrointestinal pH

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Pigs are unable to fully utilize phosphorus bound to phytate. Therefore, phytase supplements on the diet improve digestion, absorption of minerals, and animals' growth. This study aimed to assess how phytase impacts the concentration of short-chain fatty acids (SCFAs) and the pH of the gastrointestinal tract in weaned piglets. A total of 126 piglets, with 6.53 ± 1.29 kg and 21 days of age, were used in a randomized block design experiment with seven repetitions and three animals per pen. The treatments evaluated were: PC) basal diet; NC) basal diet with reduced minerals, proteins, and amino acids; T1, T2, T3 and T4) NC + 500, 1,000, 1,500 and 2,000 FTU/kg of diet, respectively. After 48 days of experiment, 42 selected animals were slaughtered to determine SCFAs concentration in cecum content, and pH in stomach, jejunum, cecum, and colon content. All data was analyzed through linear regression. Also, the NC was compared with each treatment using the Dunnett's test. For the SCFAs, there was a linear decrease for treatments with higher dosages of phytase ($P < 0.05$). Lower concentrations of propionic acid were found among animals fed with treatments T1, T2, and T4 vs NC ($P < 0.01$); butyric acid among dietary treatments T2 and T4 vs NC ($P = 0.01$); and valeric acid among all treatment's vs NC ($P < 0.01$). In terms of content pH, treatments PC, T2, T3, and T4 showed higher jejunum pH compared to NC ($P < 0.01$). Additionally, there was a trend towards increasing cecum content pH for the diet with higher doses of phytase ($P = 0.058$). Results suggest that elevated phytase doses in the diet of weaned piglets can reduce SCFAs production, raising pH levels in the jejunum and cecum, suggesting an improved intestinal environment, potentially impacting the metabolism of nursery pigs.

In ovo postbiotic stimulation of broiler chicken and impact on intestinal health

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The gut microbiota plays a key role in keeping poultry healthy. It affects the host's organism by regulating the immune response, metabolic and digestive processes, and the absorption of nutrients. The substances that might actively support rebuilding and enhancing the intestinal microbiota are bioactive components such as prebiotics or postbiotics delivered in ovo. These bioactives may eliminate intestinal dysbiosis, improve intestinal barrier and immune system. The peri-hatching period is crucial for programming the microbiota to enable colonization of the embryo's intestines with beneficial bacteria before hatching. The aim of the study is to analyze the effect of administering sodium butyrate (SB) during egg incubation on the intestinal microbiological profile and mucosal response. The in ovo injection into the egg air chamber was performed on day 12 of incubation. 1000 ROSS 308 eggs were divided into groups: control injected with saline, SB 0.1%, SB 0.3%, and SB 0.5%. The chickens were slaughtered on day 42 of rearing, and the cecal content and mucosa were collected. The relative abundance of indicator bacteria was determined using the qPCR method. RT-qPCR determined gene expression for the immunological panel (Il-1 β , Il-2, Il-4, Il-6, Il-8, Il-10, Il-12, IFN γ , AvBD1) and a panel related to the intestinal barrier function (CATHL, TJAP, CATHL2, MUC2, MUC6). SB in each dose significantly reduced the abundance of *Lactobacillus* spp. Adding 0.3% affected the promotion of *Bifidobacterium* spp. and *Faecalibacterium prausnitzii*. An increase in the abundance of *F. prausnitzii* was also determined at a dose of 0.1%. The analysis of relative gene expression showed a significant increase in the level of Il-1 β (SB 0.3%), Il-8 (SB 0.1%), and MUC2 (SB 0.1% and 0.3%). The lowest dose of SB resulted in a lower relative MUC6 level. Lower doses of SB may positively affect the bacterial profile of the cecum. The lower dose of SB resulted in a greater response from the cecal mucosa. This was reflected in activating some pro-inflammatory factors and down-regulating the anti-inflammatory response. The study was financed by grant UMO-2021/43/D/NZ9/01548 funded by the National Science Centre (Poland).

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Poster 23

Physiological response of piglets at weaning according to daily growth during lactation

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A study was carried out with 100 Duroc x (Landrace x Large White) newborn piglets, from 25 sows (4 per sow) belong to a same batch, to evaluate the effect of average daily gain during lactation (ADGlact) on some blood biochemical parameters at weaning. Piglets were weighed at birth and at weaning (1 and 21 d of age, respectively). Two equilibrate groups of piglets were classified according to ADGlact; high (124.6 \pm 32.65g/d of mean) vs low (214.7 \pm 31.57g/d of mean). No adoption was carried out. Creep-feeding was given from 7 d of age. From each piglet, a blood sample by jugular venipuncture was taken at weaning day and serum was analyzed for: sodium (Na), potassium (K), urea, creatinine, GOT (glutamic-oxaloacetic transaminase), GPT (glutamic-oxaloacetic transaminase), occludin, TNF- α (tumour necrosis factor- α) and ROS (reactive oxidative species). Data were analyzed by SAS package. As it was expected, the lower ADGlact produced lighter piglets at weaning (4.15 vs 6.26 kg; P<0.0001). In addition, those piglets with lower ADGlact had higher Na/K ratio (P=0.04) and higher levels of urea (P=0.002), occludin (P=0.007) and TNF- α (P=0.02) and lower contents of GOT (P=0.005), GPT (P=0.04) and ROS (P=0.01) than those with higher ADGlact. Pearson's correlation matrix showed that, ADGlact has (P<0.005) an important positive relationship with creatinine, GOT and ROS values meanwhile has opposite situation with Na/K, urea, occludin and TNF α . In conclusion, ADGlact had great influence on physiological response of piglets at weaning where the nutrition has a strategic role.

The effect of diets containing low doses of antibiotics on the growth and slaughter performance of broiler chickens

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The study aimed to evaluate the effect of diets containing low doses of antibiotics, simulating cross-contamination of non-medicated feed, on the growth and slaughter performance of broiler chickens. One-day-old broilers were divided into seven groups, each comprising 6 replicates with 10 birds per replicate. Until 35 days of age, the birds were fed iso-protein and isoenergetic diets containing colistin, doxycycline, flumequine, thiamphenicol, thiamulin, tilmicosin (groups I to VI, respectively) at 1% of recommended concentrations, or antibiotic-free diets (group VII). Growth performance parameters were monitored and calculated weekly, and slaughter analysis was conducted at 35 days of age. The results were analysed by one-way analysis of variance and Duncan's test. During the first week of life, birds receiving diets containing colistin, flumequine, and tilmicosin exhibited significantly higher feed intake and poorer feed conversion ratios compared to the control group, which demonstrated the most favorable FCR. Additionally, birds receiving flumequine showed the lowest body weight gains at 1-7, 15-21, and 29-35 days of age. Moreover, a significant decline in feed utilization was observed in chicks receiving colistin during the third week of life. Despite these differences in growth parameters, slaughter analysis revealed no significant variations in slaughter performance or basic carcass element percentages. In conclusion, exposure to low doses of antibiotics affects the growth parameters of chickens in a varied manner without impacting slaughter parameters, with flumequine notably negatively influencing growth parameters. This research was funded in whole by the National Science Centre, Poland, under project no 2021/41/B/NZ9/04114.

Predicting fillet lipid composition of *Oncorhynchus mykiss* using different statistical tools

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Lipid composition of animal products is pivotal for human health, seafoods are not an exception. Objective of this study was to benchmark different sources of information and statistical tools to predict fillet lipid composition in rainbow trout (*Oncorhynchus mykiss*). The fatty acids (FA) profile of aquafeeds and fish flesh were determined by gas-chromatography. A total of 228 records on single-fish analyses was available. Fish were fed 25 different diets and housed in 77 tanks. The dataset was assembled from five trials. Fish diets were characterized for their chemical composition, ingredient proportion as well as FA composition, mainly docosahexaenoic (DHA, C22:6n-3), eicosapentaenoic (EPA, C20:5n-3) acids, the total of polyunsaturated FA omega-3 (n-3 PUFA) and the sum of EPA+DHA. These sets of variables were used as predictors. Statistical models included multiple linear regression with shrinkage (MLR), partial least square regression (PLS) and random forest (RF). A three-fold cross-validation was set up, training on 2 fish samples from each tank and validating on the remaining samples. Pearson correlation between predicted and measured values in the validation set was used as a measure of prediction accuracy, averaged over the three replicates. Results show predictions based on the proportion of ingredients and the diet's chemical composition achieved accuracy of at least 0.75 for the studied fillet characteristics. As an alternative, the diet's lipid composition can be used alone. The use of all sets of predictors in the same model seemed to be redundant. Random forest outperformed the other statistical tools, suggesting that the predictors act in interaction rather than marginally. Results are encouraging and the quality of trout fillet could be predicted leveraging on diet description, with some degree of accuracy. Further studies will focus on assessing the predictive ability of the same models when a specific diet composition is not being studied and yet included in the training set.

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<i>Le Luel, M.</i>	629	<i>Lemański, A.</i>	745
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<i>Legris, M.</i>	868	<i>Li, F.</i>	532, 596, 715, 735, 971
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<i>Rodriguez Navarro, A.B.</i>	912	<i>Rossignol, M.N.</i>	658
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