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Book of Abstracts

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Continuous culture fermenters (CCF) are often used to study rumen fermentation in vitro as an alternative to in vivo trials. Here we have compared the characteristics of the fermentation liquid (FL) from a CCF with the correspondent rumen inoculum (RI). The RI was collected at the slaughterhouse from 24 bulls (8 animals in each of three separate sessions) and used as inoculum for the CCF. The diet fed to bulls was used as a substrate for the fermenters and FL samples were collected on day 6, 7 and 8 of fermentation. RI and FL samples were analyzed for VFA concentration and 16S rDNA amplicon sequencing was used to characterize the bacterial population. Total VFA concentration was lower in FL compared to RI (56.6 *vs* 114.6 mM, $p < .01$). Propionate, butyrate and isovalerate were higher ($p < .01$, 0.05 and 0.01, respectively) in the FL compared to the RI (26.6 *vs* 19.2, 13.1 *vs* 11.3 and 3.45 *vs* 1.38 mol/100mol, respectively), while acetate was lower (55.3 *vs* 66.6 mol/100mol, $p < .01$). Variability in VFA content was estimated by the error (E) from a statistical analysis, which considered the type of fluid within the session. The molar percentage of propionate, isobutyric, butyric, isovaleric and total VFA in the FL had E values (13, 18, 11, 19 and 9%, respectively) lower than for RI (17, 25, 16, 45 and 15%, respectively), while similar values were obtained for acetate (5%). Ion Torrent 16S rDNA amplicon sequencing generated 2.57M high-quality sequences clustered into 4,918 unique OTU's with 11,199 sequences per sample after normalization. At the Phylum level, the *Firmicutes* did not vary between fluids, averaging 25.1%, *Bacteroidetes* decreased by about 20% in the FL (45.0 *vs* 55.3%, $p < .01$), whereas, *Proteobacteria*, *Tenericutes* and *Spirochaetes* increased in FL compared to RI (11.3 *vs* 7.6%, $p < .05$; 2.7 *vs* 0.8 $p < .05$; 11.8 *vs* 5.0%, $p < .01$, respectively). There was no effect on *Fibrobacteres* (averaging 2.1%). Both the Shannons and Simpson index of diversity was lower in FL than in RI (4.90 *vs* 5.63, and 0.973 *vs* 0.984, $p < .01$, respectively) as was the Chao1 estimate of total species richness (1268 *vs* 1965, $p < .01$). Although, the differences in VFA concentrations and microbial populations between RI and FL were statistically significant, the studied CCF environment favored maintenance and growth of the major bacteria phyla found in RI.

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Differential gene expression analysis in broiler chickens in response to dietary larvae Mealworm meal inclusion

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Recent studies on the exploitation of larvae meal inclusion in broiler chickens diet report a significant increase of breast muscle, carcass quality and growth performance. The purpose of this study was to investigate the effects of Mealworm (*Tenebrio molitor*, TM) meal inclusion in broiler chicken diet on global gene expression of four tissues, namely breast muscle, liver, jejunum and caecum. Isonitrogenous and isoenergetic diets were formulated with 0% (control group) and 15% (test group) of TM meal inclusion. 80 one-day-old male broiler chicks (*Ross 708*) were reared in 10 pens (eight birds/pen): the pens were divided in two groups including five experimental replicates for each diet. The birds were slaughtered at 53 days of age. The tissue samples were collected and stored in RNA Later solution (Ambion) for gene expression analysis. RNA-seq was carried out on Illumina NGS analyzer. The results showed that 117, 118, 119 and 182 RNA transcripts were upregulated and 120, 116, 168 and 126 RNA transcripts were downregulated in breast muscle, liver, jejunum and cecum respectively. In all tissues 25% of downregulated differentially expressed genes (DEG) had a fold-change > four; while the 25% of upregulated DEGs had a fold-change higher than four in breast and liver and > two in intestinal mucosa. A gene ontology analysis showed that only half of DEGs were involved in known biological processes including protein metabolism (30 DEGs), gene expression (30), signal transduction (15), and immune system (21). An expression pathway analysis with Reactome showed that the pathway with minor False Discovery Rate (FDR) was the striated muscle contraction in breast and the peptide chain elongation in the other tissues. The ubiquitin B gene (*UBB*), that plays an important role in protein's metabolism, was downregulated (-0.8) in breast and upregulated (+0.57) in liver, whereas the expression of this gene did not change in intestinal mucosa. The differential expression of UBB might suggest an increase of non-lysosomal intracellular protein degradation related to a fast protein turnover in liver and a

slow protein turnover in breast. In addition, the ribosomal protein (RPLP) profile, related to peptide chain elongation, was similar: -0.45 mean fold-change in breast and +0.5 in liver while a moderated increase in intestinal mucosa was observed. No alterations in RNA expression were detected that could discourage the use of larvae mealworm inclusion in broiler chicken diet.

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Dietary supplementation with linseed and vitamin E affects sheep immune response in transition period

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The transition period represents a critical stage in sheep farming. Compared to other periods of the production cycle, transition ewes are exposed to high levels of oxidative stress, which can compromise the immunological status of the animals. If the management of the diet is not correctly performed, negative effects on health status, productivity and farm profitability can be easily observed. A number of bioactive compounds can be added to the diet to reduce oxidation and improve immune system. Linseed and vitamin E are commonly used to increase the dietary level of polyunsaturated fatty acids and decrease the oxidative stress, respectively, but few studies have investigated their effects on ewes during transition period. In the present study, 26 Sarda ewes and 22 Lacaune ewes were randomly assigned to one of the three experimental groups: 1) control feed with no supplementation (CTR), 2) CTR supplemented with 22% of extruded linseed (L), 3) CTR supplemented with 22% of extruded linseed and vitamin E (320 ppm) (LE). All concentrates were isoenergetic and isonitrogenous and were administered (from 400 g/d in late pregnancy to 600 g/d in early lactation) with alfalfa hay ad libitum. Antibody production against *Salmonella abortus ovis* after vaccination were evaluated in blood samples collected at 0, 7, 14, 21, 28 and 80 days from vaccination. To evaluate the relationship between diet and antibody production, the odds ratio (OR) analysis and the Pearson chi-square test (χ^2) were performed using the Stata 11.2 software. Significance was declared for $p < .05$. Antibody titers were grouped in two major classes: 1) "HIGH", with values greater than or equal to 1:320; and "LOW", which included values below 1:320. An influence of the dietary treatment on antibody production was detected at 14 days after vaccination: both LE (OR = 14.4, $p < .001$) and L titers (OR = 5.4, $p < .05$) were higher than those observed in the CTR group. The difference between CTR and LE group was confirmed when analyzing data with the breed included in the model (Lacaune: OR = 15, $p < .05$; Sarda: OR = 13.33, $p < .05$). The present study shows interesting results concerning the immune state of dairy ewes in the transition period, but further studies are required to better understand the relationships among oxidative status, metabolic pathways and animal welfare.

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TNF-α and adiponectin evolution in lactating ewes and goats and interactions with dietary carbohydrates

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In lactating animals, TNF-α has been associated with reduced galactopoietic activity of growth hormone, increased body fat deposition, and reduced adiponectin concentration. This work assessed possible differences between ewes and goats in TNF-α and adiponectin and their interaction with the type of dietary carbohydrate (starch or highly digestible fiber) used during the lactation.

Twenty-four healthy Sarda ewes and 24 healthy Saanen goats were compared in early and mid-late lactation. After parturition, both species were fed a high starch diet (20.4% starch, 35.5% NDF, 16.2% CP, DM basis). At 52 ± 3 DIM, 8 ewes and 8 goats were slaughtered. At 92 ± 11 DIM, the remaining 16 ewes and 16 goats were divided in two subgroups per species and fed either a high starch diet (HS: 20.0% starch, 36.7% NDF, 15.5% CP, on DM) or a low starch-high digestible fiber diet (LS: 7.8% starch, 48.8% NDF, 15.6% CP, on DM), obtained by partially replacing cereal grains with soyhulls. At

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