XIX Congress of European Mycologists
Perugia, September 4th-8th, 2023

Book of Abstracts

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XIX Congress of European Mycologists, Perugia, September 4th-8th, 2023

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Greetings and Wishes by the Honorary Members

Prof. Maria Angelova

Dear colleagues,
Dear friends,

I would like to thank the Organizing Committee for the opportunity to be a part of the big family of European mycologists. At the present meeting, I believe that many important topics will be discussed.

I wish to draw your attention to oxidative stress, one of the key phenomena of the 21st century. As eukaryotes, fungi are a convenient model to study cellular stress response. The resulting inferences can be valid for the higher eukaryotes. The research on the relationship between oxidative stress and cold exposure in Antarctic fungi is an important direction. Today we know that cold stress provokes increased generation of free oxy-radicals, accompanied by increased levels of stress biomarkers, activity of antioxidant enzymes, and expression of the genes responsible for these enzymes. At the metabolic level, many changes in the activity of the glycolytic pathway and TCA cycle were detected.

I strongly encourage young mycologists to take this research to the next level. The implementation of advanced OMICs technologies and CRISPR/Cas systems may provide a more comprehensive understanding of adaptation to cold-induced oxidative stress and survival in cold habitats.

In closing, I sincerely wish you a successful and productive forum!

Prof. Paola Bonfante

Dear colleagues,
Dear friends,

Microscopic entities or giants, friends or killers, degraders or producers, fungi are everywhere: soil, air, water. Fungi play crucial roles for our planet and for all its inhabitants, from algae to plants and animals, including humans. As in the poem *Mushrooms* by Sylvia Plath, they are a symbol of soft power and determination. Even if most of them are not visible, they produce an impressive biomass, around 12 gigatons of carbon, a figure which locates them immediately after plants and bacteria, and confirms their relevance for the structure and dynamics of the biosphere. By positively or negatively impacting on plants nutrition and photosynthesis, fungi are ultimately at the basis of the Earth food webs, with a strong influence on crop productivity and food safety.

We do not know how many fungi are still there, waiting to be discovered. Among what is left to be done is a better understanding of their diversity (also by sequencing their genomes), and of how their activities may be impacted by climate change. We should also aim at deciphering the molecular basis of their impressive ability to exploit a large variety of substrates.

As researchers and mycologists, we have the duty to bring fungi at center stage of scientific communication and to highlight to society their importance for the balance of our planet.

Prof. Silvano Onofri

Dear colleagues,
Dear friends,

To be a mycologist. The passion to walk in the wood since very young, searching and trying to recognizing fungi; the chance to have a great success in the Mycology examination at the University, resulting in the offer from the professor to attend the Mycology Laboratory at the University of Rome for internship. The insatiable curiosity of knowing the name of the encountered organism, its role in the ecosystem, its functioning and limits of life. The difficulties encountered to be accepted within international projects, as an expert of microfungi: they are not part of microbiology or botany! The long friendship with Walter Gams, my Mentor and Teacher for taxonomy and systematics of fungi. The great opportunity to visit and exploring Antarctic mycology, the best way to understand the extraordinary capability of resistance and resilience of the extraordinary Kingdom of Fungi. The chance to meet and work with E. Imre Friedmann and Gerda Hornek, discovering the cryptoendolithic world and the possibility to study the microbiology in space, until the International Space Station, demonstrating that Antarctic microfungi could survive in space and on Mars, possibly actively metabolizing in the martian subsurface. To be proud to know that nowadays Mycology is a well-respected (and funded!) independent science, with large unknown fields, deserving to be explored.

Beatrice Senn-Irlet

Dear mycologists, dear friends

It gives me a great pleasure to be able to meet you all in person again. What is mycology, what is science without meetings, without congresses? On the one hand, the individual research can be presented to a wide audience and even if the results have already been published, the personal presentation is always much more impressive and unforgettable because the researcher is visible in all his personality. Yes, also in science the context is often important. The results of modern, often rather abstract research become more approachable.

The personal exchange of ideas is immensely important, even apart from the lectures. It is only in the mutual exchange of ideas that other views become more easily understandable and new questions emerge which may lead to new joint projects.

Congresses are always opportunities to meet old friends. They always give us new impetus to move forward. We are a family!

And it is always a pleasure to welcome young mycologists. They will carry our field forward.

I am looking forward to exciting lectures and poster sessions to get to know many new aspects of mycology. The topic of fungi is currently experiencing a real hype (keyword: Fantastic Fungi), the underground network is fascinating, the biology of these organisms is fascinating. Fungi are even incorporated in revised national constitutions.

KEYNOTE LECTURES*

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* Title sorted alphabetically according to the Congress program, in [ ] number of topic.

Current perspectives, progress, evidence and challenges on medicinal mushrooms in Europe [Topic 4]

Giuseppe Venturella[1,2]

[1]Department of Agricultural, Food and Forest Sciences, University of Palermo, Viale delle Scienze, Bldg. 5, 90128 Palermo, Italy; [2]NBFC, National Biodiversity Future Center, Piazza Marina 61 (c/o palazzo Steri), I-90133 Palermo, giuseppe.venturella@unipa.it

Mushrooms are appreciated for their culinary and nutritional value and are increasingly valued for their medicinal properties, especially for their activities on the immune system, hypocholesterolemic agents and, as anticancer drugs.

Mushrooms are well known to the scientific community for having different health benefits and exhibit a wide range of pharmacological activities, including lipid-lowering,antihypertensive, antidiabetic, antimicrobic, antiallergic, anti-inflammatory, anticancer, immunomodulating, neuroprotective and osteoprotective actions.

Penicillin, lovastatin, and other globally significant medicines have been obtained from fungi, but many other species constitute poorly exploited resources from an industrial perspective. In fact, more than 90% of fungal species have never been screened for antibiotics or other useful compounds.

With the organization of the first International Medicinal Mushroom Conference (IMMC), held in Kiev (Ukraine) in 2001 the term "Medicinal Mushrooooms" was accepted worldwide. According to Lindequist's definition medicinal mushrooms are “macroscopic fungi, mostly higher Basidiomycetes, which are used in the form of extracts or powder for prevention, alleviation, or healing of diseases and/or for nutritional reasons”.

European countries boasts a long tradition in mycological studies but the interest in medicinal mushrooms has grown progressively only in the last decade, although the first example of application on humans dates

of lung metastases, inflammation and oxidative stress.

The medicinal mushroom studies in Europe are mainly characterized by four species: *Ganoderma lucidum, Cordyceps sinensis, Inonotus obliquus, Hericium erinaceum* , and *Auricularia auricular-judae*.

These fungi are overstudied in the literature, and there is a need to expand the number of species of potential application interest. Remarkable studies emerge from the activities of various researchers who have focused their attention on other taxonomic groups and or species such as *Fomitopsis officinalis, Boletus edulis, Tricholosporum goniospermum,* and the *Pleurotus eryngii* species complex.

In Europe, medicinal mushrooms are mainly marketed in the form of food supplements as single components or combined with other nutraceuticals. A huge amount of mushroom-based products is marketed in the form of powders and extracts not always of established origin and sometimes of doubtful taxonomic identification, and thus not meeting the quality criteria required.

Besides , there are failure to answer the questions regarding the safety and efficacy of medicinal mushrooms, critical issues that affect the future mushroom medicine development, that could threaten its usage in the future.

The growing interest in mycotherapy involves a strong commitment from the scientific community to propose supplements of safe origin and genetic purity as well as to promote clinical trials to evaluate its real effects on humans.

To explore the potential of medicinal mushrooms, not only as dietary supplements but also as approved drugs, increasingly intensive scientific research based on interdisciplinary collaborations, pharmaceutical industry involvement, and funding sources are needed.

Fungal communities in trees: development in space and time [13]

Lynne Boddy[1]

[1]School of Biosciences, Cardiff University, Wales, UK BoddyL@cf.ac.uk

A tree is never just a tree, it is a holobiont - a tree plus very many fungi and other microbes. These include cryptic endophytes, decomposers and pathogens of all tissues, and mycorrhizal fungi. I will concentrate on endophytes and decomposers of woody tissues. As trees age some twigs and branches cease to transport water because foliage is light suppressed or roots are damaged, likewise the central tissues in the trunks/boles stop conducting water, and become aerated allowing fungal communities to establish. The initial decay communities are largely determined by abiotic conditions in the wood and, since abiotic environment varies both between tree species and between branches and trunks, so do fungal communities. For example, central trunk tissue - heartwood - sometimes contains compunds that are inhibitory to fungi, whereas sapwood of angiosperm tree branches usually does not. The heartwood of oak (*Quercus*) is rich in tannins and other polyphenols, has a pH of around 3.5 and relatively few species, e.g. *Fistulina hepatica*, of wood decay fungi have evolved to cope with such stressful conditions. In contrast, the central tissues of beech (*Fagus*) trunks are much more benign and many more fungal species can develop. The fungi that develop initially enter in mays including through large broken limbs, damaged roots, by killing root tissues or as latent endophytes. In branches initial communities usually form from fungi latently present in functional sapwood. Although different angiosperm trees probably have a similar set of latent endophytic propagules in their sapwood different initial decay communities develop in branches of different tree species due to differnt microclimatic factors such as rate of drying, temperature and gaseous regime. Over the course of decay fungal communities change with time, driven by combative interactions as new fungi arrive, amelioration or intensification of abiotic stresses. The outcome of combative interaction are themselves influenced by environmental conditions. Since environmental conditions are changing as a result of anthropogenic activity the structutre, development and activity of decomposer communities are also likely to alter and there is already indication of changes. This talk will be illustrated largely with examples of fungal communities in attached branches and standing trunks of oak and beech trees.

Fungal materials. [5]

Han AB Wösten

[1]Microbiology, Utrecht University, Padualaan 8, 3584 CH Utrecht, The Netherlands. H.a.b.wosten@uu.nl

Pure and composite fungal materials can be part of the transition from a linear to circular economy. A palette of such materials has been produced with foam-, elastomer-, and polymer-like properties, and properties of natural materials such as wood, cork and leather. Life cycle analysis has shown that fungal materials have a much better performance than building materials and leather with respect to energy use and CO_2 emission. In fact, CO_2 is fixed in the fungal composites. So far, focus has mainly been on mechanical properties (e.g. compression strength and elasticity) and acoustic and thermal insulation. Fungal materials could exhibit other properties as well such as storage of micro-nutrients, conductivity and bioactive properties such as antibacterial or immune-stimulatory properties.

The evolution of phytopathogenic fungi. [2a]

Pedro W. Crous[1]

[1]Westerdijk Fungal Biodiversity Institute, Royal Netherlands Academy of Arts and Sciences
*Corresponding author: e-mail: p.crous@wi.knaw.nl

Key words: biodiversity, fungal evolution, genome-based prediction, lifestyles

Recent results from large-scale DNA sequencing projects have shown that most biodiversity on earth is actually very small, represented in insects and microbes. The kingdom *Fungi* forms a highly diverse, relatively unexplored lineage of eukaryotes that shares a common ancestor with animals. Both comprise heterotrophic organisms, but *Fungi* form (chitinous) cell walls and are exclusively osmotrophic; that is, nutrient uptake is extracellular. Although nearly 150,000 species of *Fungi* have been described, between 2.2 and 3.8 million are estimated to exist. Many habitats, ecosystems and host plants have, however, never been investigated, and thus their microbial inhabitants remain unexplored, unknown, and underutilised. Over the past 10 years, mycologists have on average described 2000 species per year, meaning that it will take more than 1000 years to simply describe the number of fungal taxa we estimate to occur on earth. Phytopathogenic fungi are important agents of plant disease, resulting in major annual losses to agricultural and forestry industries. The *Dothideomycetes* and *Sordariomycetes* are two of the largest and most diverse classes of ascomycete fungi, with thousands of phytopathogenic species comprising an incredible diversity of lifestyles, many of which have evolved multiple times. Studying their evolution has significant implications for our fundamental understanding of fungal evolution, and practical implications regarding the effects of climate change on these pathogens in agriculture. The availability of whole-genome data produced a high-confidence overall phylogeny of these classes, providing a clearer picture of the relationships among the various families, indicating that pathogenicity evolved multiple times within this class. Within *Dothideomycetes*, ancestral character state analyses support a terrestrial saprobic lifestyle as being ancestral within the class, also at ordinal and family levels, and that several transitions have occurred to evolve lichenised, plant and human parasitic, ectophytic (sooty blotch and flyspeck) and more recently epiphytic (sooty mould) lifestyles.

ORAL PRESENTATIONS*

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* Title sorted alphabetically according to the Congress program..

A class-wide genomic tendency throughout specific extremes in Black Fungi.[3]

[1]Laura Selbmann., [2]Sybren de Hoog, [3]Tania Kurbessoian, [3]Jason E. Stajich, [1]Claudia Coleine

[1]Department of Ecological and Biological Sciences, University of Tuscia, Italy, Viterbo, Italy
[2]Radboud University Medical Center, Nijmegen, The Netherlands
[3]Department of Microbiology and Plant Pathology, University of California, Riverside, Riverside, California, USA.

Black fungi compose a polyphyletic and morpho-ecological group within Ascomycota, mainly in the classes Eurotiomycetes and Dothideomycetes, and account among the most successful extreme-tolerant organisms on Earth. In fact, the group as a whole displays an exceptional skill to exploit all kinds of extremes, spanning from hypersaline and acidic sites, toxic hydrocarbon-contaminated environments, glaciers, hot and cold deserts, solar panels, building roofs, and exposed rocks in Polar and high altitudes in Alpine regions (1; 2; 3; 4). Melanin-like pigments, thick cell walls, flexible morphology, and meristematic phenotypes, either after switching or as a stable character, are expressions of convergent evolution. These are adaptations facilitating persistence and diversification under stress conditions up to the edge of life, which include UV- and ionizing-radiation, heat and desiccation, toxic metals, and organic pollutants, as well as biotope switches. To date, genome sequencing of this group is still largely undersampled, hampering our understanding of the evolution and molecular adaptation strategies of this intriguing fungi. In *Eurotiomycetes,* genomes mainly belong to the single family *Herpotrichiellaceae*, and reference genomes are even less representative in *Dothideomycetes*, albeit if they comprise the majority of true extremophilic black fungi. Here, we present a comparative genomic analysis of over 100 strains dothideomycetous and eurotiomycetous black fungi, selected to represent diverse ecologies, life-styles and a global distribution for a comprehensive study of adaptations of these fungi. We focus on genomic traits associated with key metabolic competences for their extremophilic behavior such as ability to withstand cold or hot temperatures, UV radiation, efficiency in DNA repair and degradation of monoaromatic toxins and pollutants. We found that the genomes of *Dothideomycetes*, much more common in extreme cold and dry environments with high naturality, were significantly enriched in cold temperature and UV radiation resistance and in genes that encode for DNA damage repair. Moreover, mean annual temperature and isothermality were positively correlated with tolerance to high temperatures in *Dothideomycetes*, suggesting that they are potentially equipped to survive even when warmer temperatures are established under global warming. Conversely, *Eurotiomycetes*, spreading mainly in hot and human impacted and polluted sites, were particularly enriched in genes related to hot temperature tolerance and hydrocarbons degradation; they were also significantly correlated with human influence.

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A comprehensive multi-method study of the fungal taxonomic and biological diversity in a model montane area [2a]

Lidia Nicola[1], Ruzbeh Hourfari[1,2], Lorenzo Rotolo[2], Carolina Elena Girometta[1]

[1]Department of Earth and Environmental Sciences, University of Pavia, via Sant'Epifanio 14 27100 Pavia, Italy, lidia.nicola@unipv.it, carolinaelena.girometta@unipv.it, ruzbeh.hourfari01@universitadipavia.it; [2]KCS Biotech SRL, via Sempione 26 Vergiate (VA), Italy, l.rotolo@kcsbiotech.it

Fungi are essential for ecosystems functioning, contributing massively to a healthy nutrient cycling, and establishing important relationships with other organisms. Environments in the Alpine system include complex ecosystems, where biodiversity is endangered both by human activities and climate change. Studying fungal biodiversity in these environments is not an easy task, and multiple methods have been used throughout time to assess it, each one with its pros and cons. In this work, three different approaches were used to assess fungal taxonomic and biological diversity in all its complexity in a model montane area. The sampling area, managed by KCS Biotech SRL (Vergiate, VA, Italy) is located in the Alpine biogeographic region inside the municipality of Crodo (Val d'Ossola, Italy) and it has an extension of 3 ha (East/South East exposition). Given its altitude (1300 m.a.s.l,), this area is at the threshold between the montane belt and the sub-alpine one. It comprises three different environments: meadow with native species, mixed forest subject to silvicultural practices with mainly Norway spruces, and an acidic bog with no anthropic intervention. Three different approaches were used to investigate the fungal biodiversity of each sub-area: the traditional mycofloristic survey of fungal sporomes, the soil serial dilution technique for counting, isolation and identification of culturable microfungi, and, finally, the metabarcoding analysis of bulk soil fungal communities. This last analysis was performed on the ITS2 fragment of the 18S ribosomal cDNA, using the Illumina MiSeq System. The mycofloristic survey detected 58 different fungal species with sporomes, especially abundant in the forest area. Most of them were mycorrhizal symbionts (mainly the genera *Russula* Pers., *Lactarius* Pers., and *Cortinarius* (Pers.) Gray), while just a few were opportunistic necrotrophic parasites. Among the most interesting species, the extremely rare *Camarophyllopsis atrovelutina* (Romagn.) Argaud. was detected in the forest area with a single specimen. Regarding the soil serial dilution analysis, the counting of Colony Forming Units (CFU) revealed the forest as the sub-area with the highest concentration of culturable fungi ($13.4 \cdot 10^5$ CFU), while the meadow and the bog had slightly lower abundances ($9.9 \cdot 10^5$ CFU, and $2.8 \cdot 10^5$ CFU, respectively). In total, 88 different culturable fungal strains were isolated. These strains mainly belonged to the genera *Penicillium* Link, *Aspergillus* P. Micheli ex Haller, *Cladosporium* Link, *Mortierella* Coem., *Acremonium* Link, *Fusarium* Link, and *Trichoderma* Pers. The metabarcoding analysis detected 3326 fungal taxa in 27 samples, belonging to 16 different phyla. The most abundant phylum was *Basidiomycota* Whittaker ex R.T. Moore, followed by *Ascomycota* R.H. Whittaker and *Mortierellomycota* Tedersoo, Sanchez-Ramirez, Kõljalg, Bahram, M. Döring, Schigel, T.W. May, M. Ryberg & Abarenkov. The soil fungal communities in the three sub-areas had comparable richness and evenness (alpha-diversity), but a significantly different community composition (beta-diversity). In the forest area, the basidiomycetous genera of *Russula* Pers., *Laccaria* Berk. & Broome and *Amanita* Pers. were among the most abundant, fact that is matching with the mycofloristic survey. Moreover, also *Meliniomyces* Hambl. & Sigler, the ericoid mycorrhiza often associated with Norway spruce, was found as very abundant. In the meadow, on the other hand, the most abundant genus was *Hygrocybe* (Fr.) P. Kumm., typical of European montane meadows, while in the bog *Mortierella* Coem. was particularly numerous. In conclusion, this area was particularly suitable for this multi-method study, since it was circumscribed but with three distinct ecosystems, all three typical of the montane/sub-alpine environment. The use three different methods for the investigation of fungal biodiversity proved to be extremely useful, since every technique focused on a different aspect; the metabarcoding analysis provided a general overview of the whole fungal communities, the mycofloristic survey focused especially the ectomycorrhizae and the ligninolytic fungi, while the serial dilution analysis detected the saprotrophic highly sporulating microfungi The last two analyses allowed also the precious isolation of specific fungal strains found in the area. This multi-method approach is extremely valuable in the prospect of the development of a conservation plan for this private montane area in the future, with a special attention on mycological conservation and divulgation.

Anaerobic digestate: a promising growing substrate for full-scale cultivation of *Pleurotus ostreatus* [12]

Veronica Zuffi[1], Federico Puliga[1], Alessandra Zambonelli[1], Lorenzo Trincone[1], Santiago Sanchez-Cortes[2] and Ornella Francioso[1]

[1] Department of Agricultural and Food Sciences, University of Bologna, Viale G. Fanin 40-50, 40127 Bologna, Italy; federico.puliga2@unibo.it (F.P.); alessandr.zambonelli@unibo.it (A.Z.); lorenzo.trincone@studio.unibo.it (L.T.); ornella.francioso@unibo.it (O.F.)

[2] Instituto de Estructura de la Materia, Consejo Superior de Investigaciones Cientificas (IEM-CSIC), Serrano, 121, E-28006 Madrid, Spain; s.sanchez.cortes@csic.es

In the last decades, the number of studies about the valorization of agricultural by-products has strongly increased due to the growing focus on circular economy. In this context, mushrooms cultivation is rising attention for their nutritional properties and their adaptability to different growing conditions (1). Fungi have a fundamental role as decomposers of the organic matter, in particular the lignocellulosic fraction, thanks to their complex enzyme machinery. Currently, almost 60 species of saprotrophic fungi are cultivated worldwide (2). Their production is considered as low environmental impact since it requires low energetic inputs and short cultivation time. In recent years, mushroom companies are researching for new alternative substrates in order to differentiate them, reducing costs and at the same time increasing production and quality parameters. Among these, agro-wastes are generally rich in lignin, cellulose and hemicellulose; for this reason, they may represent potential growing matrices. By using these wastes, it may be possible to decrease the costs of raw materials, to valorize a waste and to increase the eco-sustainability of two production chains (3).

The goal of this study was therefore to promote a by-product, the solid anaerobic digestate fraction from biogas plant, as a potential substrate for *Pleurotus ostreatus* full-scale cultivation. A preliminary in vitro trial shows that the mycelia can colonize with the same rate both the conventional substrate (wheat straw added with sunflower cake), and the experimental ones supplemented with corn digestate. The most important and innovative part of the work was the full-scale cultivation. Quantitative analyses demonstrate that *P. ostreatus* is capable of growing on the conventional substrate as well as on the substrate containing 15% of digestate without reducing the mushroom harvest. Even better, digestate seems to stimulate a precocious and more homogeneous production. Moreover, chemical and spectroscopic analyses carried out on the fruiting bodies confirm the results; in fact, mushrooms show a comparable structural composition, highlighted with Fourier Transformed Infrared (FTIR) and Surface-enhanced Raman Scattering (SERS) spectroscopic evaluations. Lastly, the nutrient content did not change due to the addition of digestate. These promising results demonstrate that anaerobic digestate can become a resource for *P. ostreatus* cultivation without production losses and maintaining the same qualitative characteristics.

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Antimicrobial activity of *Pisolithus arhizus* methanolic extract: preliminary results against some human pathogenic bacteria [4]

Giulia Mirabile[1,2]

[1]Department of Agricultural, Food and Forest Sciences, University of Palermo, Viale delle Scienze, Bldg. 5, 90128 Palermo, Italy; [2]NBFC, National Biodiversity Future Center, Piazza Marina 61 (c/o palazzo Steri), I-90133 Palermo, giulia.mirabile@unipa.it

In recent years, there has been an increased interest in new natural antimicrobial compounds obtained by medicinal mushroom due to the serious issue of antibiotic resistance. Some bacteria such as *Klebisiella* spp., *Escherichia coli*, *Enterococcus* spp. and, *Pseudomonas* spp., have been drastically increasing their resistance to first-choice antibiotics (1). Therefore, the research of new antibiotic substances active against human pathogenic bacteria is crucial. In this regard, mushrooms extracts could be an alternative source of new antimicrobial substances (2). Recently, several studies showed the antimicrobial and medicinal potential of edible ectomycorrhizal species which seems to represent an unlimited source of bioactive compounds (3). In this study we tested a methanolic extract of *Pisolithus arhizus* (Scop.) Rauschert (Sclerodermataceae, Basidiomycota), ectomycorrhizal species of *Abies* spp., *Acacia* spp., *Betula* spp., *Pinus* spp. and other forest trees, against the pathogenic human bacteria *Escherichia coli* (Migula, 1895) Castellani & Chalmers, 1919, *Salmonella enterica* Le Minor and Popoff 1987 subsp. *enterica*, *Listeria monocytogenes* (Murray et al. 1926) Pine 1940, and *Staphylococcus aureus* subsp. *aureus* Rosenbach, 1884. To test the antimicrobial activity of fungal extract, paper disk diffusion assay was used. Subsequently, microdilution test was carried out to determine minimum inhibitory concentration (MIC) values of the extract. *Staphylococcus aureus* ATCC 33862 was found to be the most sensitive with an inhibition zone (halo around the paper disk) of 13 ± 0.1 mm, while *Escherichia coli* ATCC 25922 was found to be the less sensitive with an inhibition zone of 9 ± 0.2 mm. Moreover, high antibacterial activity (percentage of growth inhibition > 70%) was detected at MIC values of 2.5 mg/ml for *E. coli* ATCC 25922, *S. enterica* subsp. *enterica* ATCC 13076 and *S. aureus* subsp. *aureus* ATCC 33862. *L. monocytogenes* ATCC 19114 was the only bacterial strain to show high growth inhibition at MIC value of 1.25 mg/ml. These results demonstrate that methanolic extract of *P. arhizus* may contain bioactive compounds useful as potential antibacterial agents. Further investigations are needed to characterize these antimicrobial compounds and to test their efficacy against other human pathogenic bacteria.

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Ants’ infrabuccal pocket as a source of fungal novelties – a discovery of *Formicomyces* gen. nov. (*Chaetothyriales*) from *Formica polyctena* ants [2a]

Igor Siedlecki[1,2*], Marcin Piątek[3], Maria Majchrowska[2], Alicja Okrasińska[2], Magdalena Owczarek-

[1] Botanic Garden, University of Warsaw, Warsaw, Poland; [2] Institute of Evolutionary Biology, Faculty of Biology, University of Warsaw, Warsaw, Poland; [3] W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, Poland; [4] Norwegian Veterinary Institute, Ås, Norway
* igor.siedlecki@uw.edu.pl

The infrabuccal pocket is a ventral sac-like outgrowth of the hypopharynx, located in an ant’s head. This organ is believed to work as a filtering device - the particles collected while ants eat or perform grooming behavior remain inside and form a pellet in which fungal hyphae and spores are often present. However, all functions of the organ remain understudied and diversity of microorganisms present in it is still not fully known. A study on infrabuccal pockets of *Camponotus pennsylvanicus*, in which *Pencillium camponotum* and *P. infrabuccalum* were discovered, shows that such an organ could be an isolation source of previously unknown fungal species (1). While studying the mycobiota of infrabuccal pellets of *Formica polyctena* (a common European ant), we have managed to isolate two slow growing strains of “black yeasts” preliminarily identified as unknown *Chaetothyriales* representatives. The aim of this work was to determine the systematic and phylogenetic position of the isolated strains. Molecular analyses based on maximum likelihood and Bayesian inference, using sequences of two ribosomal markers: ITS and LSU, for 78 isolates, have shown that the isolated strains form a monophyletic clade within the family *Trichomeriaceae*, sister to a clade formed by representatives of the genus *Trichomerium*. Morphological analyses, based on micro- and macroscopic characteristics, additionally justified distinctiveness of the isolated strains, which have different morphology of conidia and conidiophores than *Trichomerium* representatives. Therefore, our results show that the isolated strains represent a new species within a not yet described fungal genus. Due to the strains’ isolation source and their close relatedness to a fungal strain isolated from a carton nest of *Lasius fuliginosus* ants, we propose a name *Formicomyces microglobosus* Siedlecki & Piątek for this fungus. While some representatives of *Trichomerium* - the sister clade to *Formicomyces*, are known to be associated with ants, mostly as symbionts which overgrow walls of carton nests of tropical species (2), further studies on *F. microglobosus* and its presence in the surroundings of temperate ant species are recommended, to fully understand this potential association.

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Are healthy protein-rich termite fungal crops promising as human food? Preliminary studies. [12]

Michał Kochanowski[1], Suzanne Schmidt[2], Søren Rosendahl[2], Michael Poulsen[2]

[1] University of Warsaw Botanic Garden, Aleje Ujazdowskie 4, 00-478 Warszawa
[2] Section for Ecology and Evolution, Department of Biology, University of Copenhagen

The cultivation of macrofungi can help feed the growing human population in sustainable ways if grown on organic waste from agriculture or the timber industry, that would otherwise be burned. Hitherto, the cultivation of mushrooms for culinary purposes has focused on saprotrophic species that are easy to grow, but not necessarily optimal for human consumption. Fungi of the genus *Termitomyces*, on the other hand, have evolved over 30 million years to become optimal food for farming termites. The fruiting bodies of these mushrooms could also be a nutritious food source for humans, containing all essential amino acids and more digestible protein than chicken meat. However, to date, obtaining these fungi for food has only been possible from collections from termite mounds, limiting the role these fungi can play for humans.

In this study, we tested the growth of five *Termitomyces* strains symbiotic with different genera of termites, from Côte d'Ivoire, on five different types of rich substrates (four different cereal seeds and sunflower seeds) and five different waste substrates (four types of sawdust and straw). Two different granulations of the selected substrates were also tested in the experiment. Growth experiments were conducted in glass tubes with a substrate that was inoculated with a mycelial fragment. After four weeks, the results showed no consistency between strains growth rates on the different rich substrates nor substrate granulation. Preliminary results suggest that, of the waste substrates, *Termitomyces* grows fastest on spruce (*Picea abies)* sawdust. The results of this preliminary study will help to undertake the next steps in optimising and scaling up the growth of Termitomyces to an industrial scale.

Biodiversity of microfungi on forest trees in the nature reserves of Türkiye and Belarus. [2a]

Faruk Selcuk[1], Tatsiana Shabashova[2], Darya Belomesyatseva[2], Merve Ulukapı[1]

[1]Kırsehir Ahi Evran University, Sciences and Arts Faculty, Department of Molecular Biology and Genetics. Kırşehir, TÜRKİYE, selcuk_faruk@yahoo.com
[2]V.F. Kuprevich Institute of Experimental Botany, Minsk, BELARUS, shtgby@gmail.com

Microfungi are the most important causes of plant diseases. The detailed investigation of the mycota in natural or protected forest ecosystems and the dynamics and pathogen of mycobiota of plant populations in the areas exposed to anthropogen effect is an important step in struggle to the parasites and pathogen microfungi species in both phytocenosis and mycosinusia. We conducted joint research on micromycetes during 2019-2022 in order to study and carry out the comparative analysis of ecology and biological diversity of micromycetes, in forest phytocenosis on the protected areas of Turkey and Belarus (on the example of the Kazdağı and Belavezhskaya Pushcha National Parks).

Kazdagi National Park is in the district of Edremit, in Balikesir province (Türkiye). It has a unique importance due to its biodiversity, endemism rate, phytogeographical location and the direction of the mountains. It is at the intersection of three phytogeographical regions that Mediterranean, Irano - Turanian and Euro-Siberian. Therefore, it is a very important location in terms of biodiversity.

Belovezhskaya Pushcha National Park (Belarus) is the most primitive forest of Europe, considered to be a World Heritage Site, where the coniferous forests are very close to the European broadleaf zone, and short-term freez events occur with the effect of Taiga. Both national parks are important areas due to the presence of common species and genera in terms of tree and bush species, as well as the location of coniferous forests where they interfere with broad leafs and the areas where short term frosts are effective.

During the project, field studies were carried out in the area and samples were collected from parts of trees and shrubs that form forests, such as branches, stem bark, leaves and fruits, which are infected with microfungi. As a result of the examination of the samples brought to the laboratory, 75 species were identified. Considering the trophic structures of the species, 34 species are xylotrophs. These are followed by lignotrophs with 23 species, phyllotrophs with 14 species and carpotrophs with 4 species. Xylotrophs and lignotrophs are saprotrophs and only 3 phyllotrophs display saprotrophic characters. 10 phyllotrophs and 3 carpotrophs are parasites. 1 phyllotroph is pathogenic. According to the literature review, 11 genera and 44 species of microfungi are new records for the Turkish mycobiota as a result of this research.

The National Park "Belovezhskaya Pushcha" (Belarus) is located at the junction of two geobotanical zones - European broadleaf and Eurasian coniferous forests, which determines a rare combination of woody and shrubby types of forest vegetation, a high degree of mosaic of the territory and a wealth of habitats. More than half of the area is occupied by forests, the average age of which exceeds 100 years, including 9.7% older than 180 years. The variety of tree species, the presence of large volumes of dead wood create favorable conditions for the development of fungi of different systematic groups. Of particular interest are microscopic fungi included in the consortium with the main forest-forming species - *Pinus sylvestris* L. and *Picea abies* (L.) H. Karst as well as on their accompanying species - oak, maple, hornbeam, birch, aspen, alder, hazel.

In total, more than 236 species of micromycetes were identified in the consortium of pine and spruce and their accompanying breeds, 113 species on pine and spruce, 123 species on their accompanying breeds. The taxonomic composition of microscopic fungi on them is specific at the level of associations, where the excess of different species of certain genera is a direct reflection of the ecological and trophic structure.

However, certain patterns of distribution of fungi by large taxa are observed - Ascomycota are more common in the anamorphic stage (147 species) and less common in the perfect stage (72 species). The highest species number of sac fungi is in the orders Helotiales (18 species) and Pleosporales (17 species). The main part of micromycetes belongs to hyphal fungi, typical saprotrophs, less often hemibiotrophs – 47.5%, Ascomycetes – 31.5%, and Coelomycetes – 14.7%. There are significantly fewer fungi of the Zygomycota and Basidiomycota phyla – each 2.5%, and the last in the number of species is Oomycetes – only 1.3% of the total number of species.

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Bullshit fungi - fimicolous fungal biodiversity hot spots on Heck cattle dung in Vienna. [2b]

Irmgard Krisai-Greilhuber[1], Markus Gorfer[2], Matthaeus Koncilja[3]

[1]Dept. of Botany and Biodiversity Research, University of Vienna, Rennweg 14, 1030 Wien, Austria, E-mail: irmgard.greilhuber@univie.ac.at; [2]Bioresources, Center for Health & Bioresources, AIT Austrian Institute of Technology, Konrad-Lorenz-Straße 24, 3430 Tulln, Austria, E-mail: Markus.Gorfer@ait.ac.at; [3]Roacher Str. 26, 9535 Schiefling am Wörthersee, Austria, E-mail: matthaeus.koncilja@aon.at

Few dung fungi are known from Austria. In 2021 and 2022, dung samples of Heck cattle kept natural from the Lainzer Tiergarten (Vienna) were examined for the presence of dung fungi.

More than 80 different fungal species were morphologically detected. Further ca. 50 by metabarcoding. Many could be identified to species, some to genus level. Comparative studies on intensively kept cattle revealed only about ten fungal species.

During the year, there were events that strongly influenced the fungal occurrence: an enormous insect pressure, which resulted in a noticeable reduction of the fungal species, and unusually massive fructifications of ink fungi, which literally sealed the substrate with spore powder and made it impossible for other fungi to form fruiting bodies. The dung samples were stored in transparent plastic containers with perforated lids and regular humidification and checked daily for fungal growth under a stereomicroscope. As far as the succession of fungal species is concerned, certain regularities in the appearance of fungal species could be observed in the different dung samples. Zygomycetes such as *Pilobolus crystallinus*, *P. kleinii*, *Mucor racemosus*, *Absidia corymbifera* usually appear first, often followed by asexual stages of ascomycetes such as *Arthrobotrys superba*, *Antromyces copridis* and other hyphomycetes. This is regularly followed by ascomycetes such as *Ascobolus immersus* in many individuals, *Saccobolus citrinus*, *S. ovibovinus*, *Coprotus disculus*, *C. granuliformis*, *Lasiobolus cuniculi* and *Schizothecium hispidulum* as well as various *Podospora* species. At the first macromycete appearance, basidiomycetes are then involved, represented by ink fungi, e.g. *Coprinellus hepthemerus* var. *pusillulus*, *Coprinellus heterosetulosus* and *C. brevisetulosus*.

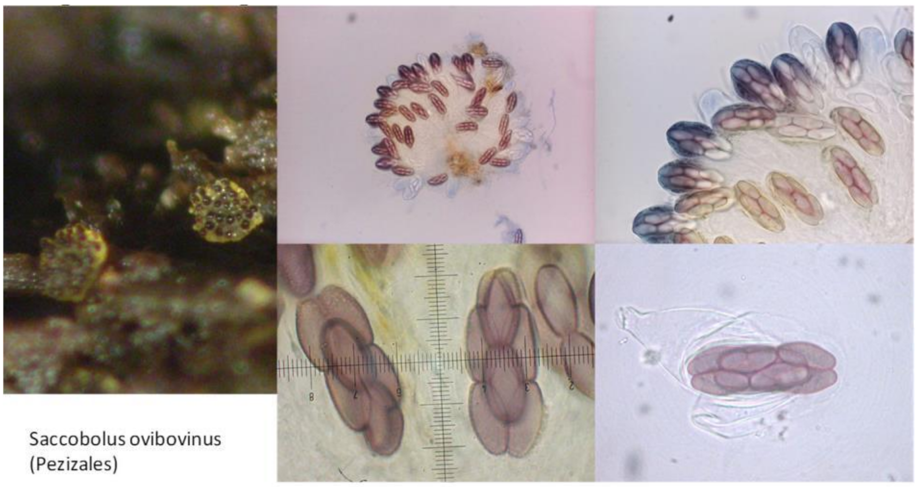

Fig. 1. Saccobolus ovibovinus recorded on dung in Austria for the first time.

The end of the fungal succession is predominantly formed by *Thecotheus pelletieri*, *Th. formosanus* f. *collariatus*, *Ascobolus hawaiiensis*, *Zygospermella insignis*, *Zygopleurage zygospora* and coprinoid species such as *Narcissea cordispora*, *Coprinellus sassii* or *Coprinopsis stercorea*. Only three species of slime moulds were found during the investigations, namely *Didymium difforme*, *Oligonema fulvum* and *Physarum leucopus*.

Parallel to the morphological investigations, some samples were also sequenced with metabarcoding. This resulted in both matches and new finds that could only be determined by sequencing. Our results show that close-to-nature cattle farming greatly promotes fungal diversity and that extensive cattle farming is very important for fungal biodiversity conservation.

Changes in soil fungi associated with New Zealand mānuka (*Leptospermum scoparium*) irrigated with treated wastewater. [7]

Sophie van Hamelsveld[1], Alexandra Meister[1], Gregory A. Hatley[1], Farideh Jamali-Behnam[1], Rawiri Smith[2], Lawrence Stephenson[3], Russleigh Parai[1], Maria J. Gutiérrez-Ginés[1]

[1] Institute of Environmental & Research (ESR), 27 Creyke Road, Christchurch, New Zealand.
[2] Ngāti Kahungunu ki Wairarapa, Aotearoa New Zealand.
[3]Carterton District Council, Carterton, New Zealand.

The majority of treated wastewater in Aotearoa (New Zealand) is currently discharged to water bodies such as the ocean. In many countries, wastewater is reused for irrigation to support the growth of plants by providing essential nutrients such as carbon and nitrogen. The New Zealand mānuka (*Leptospermum scoparium*; photo 1), is widely cultivated for mānuka honey and essential oil production. Mānuka and its relatives are important species in New Zealand forests, playing host to many endemic fungi. It is these fungi, particularly mycorrhizal species (photo 2), that may increase plant survival by providing essential nutrients. Mānuka is also a pioneer species. It is therefore important for replanted mānuka to thrive so that native forests can be regenerated. Our stakeholders were interested in how wastewater could be utilised to improve the health and quality of mānuka plants to aid in productivity and forest regeneration efforts. We therefore performed an eight-month long greenhouse trial to investigate the outcomes of different levels of treated wastewater irrigation on plant and soil health. We used amplicon and shotgun metagenomic sequencing to measure the diversity and abundance of fungi and other microorganisms in mānuka root-associated soil. We collected other applicable data such as soil nutrient content and plant biomass. We previously showed that wastewater application can increase available carbon and nitrogen in soils. Saprophytic fungi may benefit from increased carbon availability. However, the need for mānuka plants to host mycorrhizal fungi might decrease with increasing phosphorous and nitrogen availability in soil. We therefore hypothesised that treated wastewater application would result in increased diversity of saprophytic fungi, but decreased diversity of mycorrhizal fungi in the soil. I will present data regarding the abundance and diversity of fungi and other microorganisms found in mānuka soils irrigated with treated wastewater. I will also share data relating to the health and chemical analysis of both soil and mānuka plants from this trial. Our results demonstrate the ways in which soil fungi can be influenced by re-use of wastewater in the context of a circular economy.

Photo 1: *Leptospermum scoparium* (mānuka) in bloom.

Photo 2: *Cortinarius coneae,* an endemic mycorrhizal species found only under New Zealand tea tree species and classified as nationally-critical.

Circular Economy of الفطريات (Champignon, Fangasi) in Africa: Current Status and Future Prospects. [7]

Ahmed M. Abdel-Azeem[1,2]

[1]Botany and Microbiology Department, Faculty of Science, Suez Canal university, Ismailia 41522, Egypt.
[2]Department of Genetics, Faculty of Natural and Agricultural Sciences, University of the Free State, Bloemfontein 9300, Republic of South Africa.

Egypt is considered the cradle of mycology where ancient Egyptians (5623 B.C.) documented the use of fungi on walls and pillars of temples, within hieroglyphic texts, ear studs and medical prescriptions. Ancient Egyptians believed that some mushrooms were plants of immortality and had magical powers e.g.

Psilocybe cubensis which documented in Hathor temple in Dendera. Later, desert truffles (*Terfezia*, *Tirmania* and *Phaeangium*) used by ancient Egyptians and in the Middle East were mentioned in the Bible and in Hadith by Prophet Muhammad peace, be upon him in the seventh century.

In Africa, edible fungi have been mainly ignored and underutilised. Not only is the number of edible species unknown, but their essential role in human food and nutrition, as well as the ecosystem services they provide & conservation, are inadequately understood and undervalued.

Scientific information about biodiversity distribution is indispensable for nature conservation and sustainable management of natural resources. For several groups of animals and plants, such data are available, but for fungi, especially in Africa, they are mostly missing. In 2023, a total number of 480 species of wild edible mushrooms belonging to 126 genera and 60 fungal families across Africa were recorded through literature. Ecologically, a total of 249 species were mycorrhizal, 28 species were termitophilic and 203 species were saprobic or parasitic.

We would assume that an industry valued at $993 billion in 2022 and projected to reach $1.7 trillion by 2030 would dispose of waste responsibly or recycle it. Unfortunately, the majority of our wastes is burned or interred underground. The improper management of waste causes more than 400 thousand deaths annually and has negative effects on the environment and adjacent communities.

Fungi can transform organic materials into a diverse array of useful products and offer unique opportunities for addressing the pressing challenges facing humanity. Myco-biotechnology can expedite the transition from our petroleum-based economy to a bio-based circular economy and has the capacity to produce resilient sources of food, feed, chemicals, fuels, textiles, and materials for the construction, automotive, and transportation industries, and beyond, in a sustainable manner. Through the rational improvement of new and existing fungal cell factories, fungal biotechnology has the potential to contribute significantly to climate change mitigation and the attainment of the United Nations' sustainable development objectives.

Recently, the Africa Circular Economy Facility (**ACEF**), a multi-donor grants trust fund hosted by the Climate Change and Green Growth Department of the African Development Bank has launched. Its objective is mainstreaming the circular economy as an inclusive green growth strategy to help African nations fulfil their development priorities while meeting the goals of the Paris Agreement, Sustainable Development Goals (SDGs) and the African Union’s Agenda 2063.

The presentation highlights the major issues and challenges that may hamper African economic growth and development and African fungi’s role and importance in addressing these to build a sustainable green economy in Africa.

Climate drives diversity patterns of dominant microbial taxa in caves worldwide. [10]

Federico Biagioli[1], Claudia Coleine[2,*], Manuel Delgado-Baquerizo[3,*], Youzhi Feng[4], Cesareo Saiz-Jimenez[5], Laura Selbmann[6]

[1]Department of Ecological and Biological Sciences, University of Tuscia; 01100, Viterbo, Italy; fbiagioli@unitus.it
[2]Department of Ecological and Biological Sciences, University of Tuscia; 01100, Viterbo, Italy; coleine@unitus.it
[3]Laboratorio de Biodiversidad y Funcionamiento Ecosistémico (BioFunLab); Instituto de Recursos Naturales y Agrobiología de Sevilla (IRNAS), CSIC, Av. Reina Mercedes 10, E-41012, Sevilla, Spain; m.delgadobaquerizo@gmail.com
[4]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science; Chinese Academy of Sciences, 210006, Nanjing, China; fyouzhi@hotmail.com
[5]Instituto de Recursos Naturales y Agrobiología de Sevilla (IRNAS), CSIC; Av. Reina Mercedes 10, E-41012, Sevilla, Spain; saiz@irnase.csic.es
[6]Department of Ecological and Biological Sciences, University of Tuscia; 01100, Viterbo, Italy; selbmann@unitus.it

To date, new uncertainty about long-term conservation of the astounding natural and cultural heritage of caves has spawned by the impending environmental shifts driven by climate change.

Although the understanding of ecology and environmental factors driving structural and spatial patterns of microbial communities represent a crucial task to assess the integrity and functionality of cave ecosystems, the influence of climate in explaining the global distribution of cave microbiomes remains virtually unknown. To address this knowledge gap, we put together over 1,050 bacterial and fungal communities of caves worldwide, coupled with a wide plethora of environmental data.

As major results we found that: i) fungi and bacteria showed different ecological preferences when in temperate-tropical and temperate-continental climatic regions, respectively; while arid climate leads to a reduction in total cave microbial diversity. ii) Despite the broad environmental and ecological variability, we clearly found a shared microbial core across caves worldwide, for the first time. iii) Climatic variation represents one of the main epistatic drivers in determining the assembling of microbial community cores. In conclusion, our work underlines how terrestrial caves and their highly adapted microbiota represent excellent natural benchmarks for the biogeographic information for caves globally and for protection strategies aiming at conservation of underground environments.

Combination of hosts and geographic origin affect the differentiation in *Fomes fomentarius* species complex. [2a]

Robert Logar[1,2], Cene Gostinčar[2], Martina Turk[2], Tine Grebenc[1]

[1]Slovenian Forestry Institute, Večna pot 2, Ljubljana, Slovenia. rl12691@student.uni-lj.si
[2]University of Ljubljana, Biotechnical Faculty, Jamnikarjeva 101, 1000 Ljubljana, Slovenia.

The genus *Fomes*, has a diverse range of hosts, primarily broadleaved trees. Although there are morphological differences among collections, significant differences in spores or other microscopic structures are limited. Molecular analyses have revealed distinct evolutionary lineages and the presence of cryptic species within *Fomes fomentarius*. Previous research and our preliminary data indicate the influence of the host tree on the phylogenetic distances in the genus *Fomes*, with one lineage associated with *Betula*, and the other with several other broadleaves such as *Acer*, *Quercus*, and *Tilia*. Despite its prevalence, little is known about the ecology, phylogeography, and host adaptations of the genus. This study aims to provide insights into the phylogenetic relationships within the genus Fomes based on comprehensive ITS sequence data. Therefore, a large-scale host sampling and phylogenetic analysis was conducted to investigate the potential role of host and geographic distance in *Fomes* spp.. In this study, sporocarps from the genus *Fomes* were collected in years 2010 and 2023 using purposive sampling to target a diverse range of potential hosts and geographic areas. Sporocarps were sampled for DNA extraction, and the nuclear ribosomal DNA internal transcribed spacer (nr rDNA ITS) region was sequenced. Additionally, all existing sequences and associated metadata belonging to the genus *Fomes* were obtained from the International Nucleotide Sequence Database (GenBank). Total DNA was extracted from sporocarps or from mycelium isolated in pure culture using a DNeasy Plant Mini Kit, and the ITS region was amplified with the ITS 1f and ITS 4 primer pair. The amplified DNA fragments were purified and sequenced at a commercial laboratory (Macrogen Inc., Seoul, South Korea). Sequences were aligned in MAFFT (E-INS-i aligning strategy), a phylogenetic network was reconstructed through the Median Joining function. Our findings indicates that host species/genus groups enforce differentiation within the *Fomes fomentarius* species complex. We identified two distinct clades within the phylogenetic tree. The first clade primarily consists of host species such as *Betula* and *Fagus*. In the second clade, the dominant host species are *Tilia*, *Populus*, *Carpinus*, and *Quercus*. From phylogenetic network the "*Fomes fomentarious*" group is relatively distant from nearest available genotype by 54 mutation events, while the two groups within the complex are separated by 18 mutation events. Genotypes within each group are separated by no more than 4 mutation events with no clear further corelation with host tree species. The length of branches between both clades suggests relatively low genetic divergence among the "*Fomes fomentarious*", indicating their close evolutionary relationship. We conclude that the host tree and potentially their postglacial migration routes have an influence on the genetic diversity of individuals within the *Fomes fomentarious* species complex and no additional cryptic species were detected from the area of SE Europe.

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Combined impact of forest management and climate change on *Boletus edulis* productivity: may mycosilviculture mitigate the effects of climate extremes?

Elena Salerni[1], Luca Paoli[2], Claudia Perini[1]

Department of Life Sciences, University of Siena, Via Mattioli 4, 53100 Siena, Italy. elena.salerni@unisi.it ; claudia.perini@unisi.it; [2] Department of Biology, University of Pisa, Via Ghini 13, 56126 Pisa, Italy. luca.paoli@unipi.it

Climate change can affect forest ecosystems, especially through an increase of extreme events. In order to verify whether mycosilvicultural practices could mitigate the effects of climate change, productivity of Boletus edulis in Abies alba managed plantations was correlated to intense rainfall and temperature peaks during three years. Fungal productivity in each of the 21 days following an extreme climatic event was considered. Results showed that sudden increases in maximum temperature seemed to have an inhibitory effect on B. edulis productivity in sites with no or modest thinning. In sites with heavy thinning, productivity seemed to be favoured by high temperatures, starting from the twentieth day following the extreme event. Mycosilviculture may mitigate the climate change effects; however, further studies are needed to verify how climate-dominated effects can be altered by local factors and how ecological relationship between B. edulis and forest ecosystem will be affected.

Community composition of wood-inhabiting fungi in Mediterranean deciduous oak woods.

Claudia Perini [1,2], Ángel Ponce[3,4], Maria Nives D’Aguanno[5], Elena Salerni[1],

[1]Department of Life Sciences, University of Siena, via P.A. Mattioli 4, 53100 Siena, Italy
[2]NBFC, National Biodiversity Future Center, 90133 Palermo, Italy.
[3]Department of Agricultural and Forest Sciences and Engineering, University of Lleida, Av. Alcalde Rovira Roure 191, E-25198 Lleida, Spain
[4]Joint Research Unit CTFC – AGROTECNIO – CERCA, Av. Alcalde Rovira Roure 191, E-25198 Lleida, Spain
[5] Institute of Microbiome and Applied Sciences, Malta Life Sciences Park (LS2.1.12 – LS2.1.15), San Gwann, SGN 3000, Malta.

Deadwood plays various crucial ecological roles among them it serves as the fundamental habitat for wood-decay fungi. Simultaneously, this polyphyletic group of fungi acts as the primary agent of wood decomposition, regulating the carbon cycle and providing a food resource for numerous other organisms. The diversity and community composition of wood-decay fungi are known to be influenced by factors such as tree species, decay stage, and size of the woody debris they inhabit. However, there is a lack of information regarding Mediterranean Turkey oak woods. In response, we conducted a study to explore how wood-decay fungi colonize different features of deadwood and how the community composition is influenced by decay stage and size of the woody debris. Our results indicate that the studied groups, namely Ascomycetes, Corticioids, Polyporoids, and Heterobasidiomycetes, respond differently to the characteristics of woody debris. Moreover, we highlight the significant importance of smaller and soft-decayed woody debris for the fungal community, as it harbours a wide range of species. Additionally, we emphasize the positive effect of the heterogeneity in woody debris size on wood-decay fungi fructification.

Comprehensive Technologies for Large Scale Medicinal Mushroom Biomass Cultivation.

Prof. Dr. Marin Berovic

Faculty of Chemistry and Chemical Technology, University of Ljubljana, Slovenia

Great interest for large scale production of various medicinal mushroom pharmaceutically active compounds requests the development of new comprehensive technologies. Production of fungal fruit bodies using farming technology could hardly meet the demand of World market.

Comprehensive large scale medicinal fungi biomass industrial production is focused in consumption of various secondary lignocellulosic wastes and low energy consumption.

Submerged liquid substrate cultivation **represents fastest and the most suitable technology for a large scale production of medicinal mushroom biomass and its pharmaceutically active compounds for human use. It** is the most suitable technology for high amount polysaccharide production. It enables high sterility and well controlled but on the other side by significant rheology changes and as a consequence limitation in mass and heat transfer also high energy consumption process.

Solid state cultivation of various medicinal mushroom is very close to fungal natural growth. In last three decades solid state cultivation of various kinds of medicinal mushroom mycelia in various types of bioreactors was also established. This technology also represents well controlled process suitable for medium scale recycling organic waste materials and for a production of valuable veterinary use products. As the substrates various secondary wastes from wood, agriculture and fruit industry are successfully used. No fungal fruit boddies are produced. Final product represents delignolized, wooden material overgrown by medicinal fungi biomass enreach with proteins and various pharmaceutically products. Dryed and pulverized it could be directly used as a special veterinary remedies in a veterinary need.

This **presentation is an overview of the engineering achievements in submerged and solid state cultivation in bioreactors.**

Data or species?

Vincent Robert[1], Gianluigi Cardinali[2], and ChatGPT OpenAI[3]

[1]*BioAware, Hannut, Belgium, vrobert@bio-aware.com*
[2]*University of Perugia, Dept. Pharmaceutical Science, Perugia, Italy, Gianluigi.cardinali@unipg.it*
[3]*OpenAI, San Francisco, California, United States, www.openai.com*

Species names are widely used in biology, ecology, biotechnology and the medical field for example but they have serious limitations and drawbacks. They are often ambiguous, inconsistent, or inaccurate, and, often, they do not reflect the genetic diversity and evolutionary history of organisms. Data, on the other hand, are more objective, reliable, and informative. Data can capture the variation and complexity of life forms and reveal their relationships and patterns. In this presentation, we argue that data should be used as the primary unit of analysis and communication in scientific studies, rather than species names. We provide a few examples of how data can be used to address various research questions and challenges, such as identifying cryptic species, reconstructing phylogenies, assessing biodiversity, and monitoring conservation status. We also discuss the benefits and challenges of using data instead of species names, and propose some recommendations for data collection, management, and sharing. We conclude that data are more powerful and useful than species names, and that using data can advance our understanding and appreciation of the natural world.

Cultivation trials of medicinal mushrooms on crop residues for sustainability of wineries.

Gaetano Balenzano[1], Valeria Ferraro[2] , Maria Letizia Gargano[1], Fortunato Cirlincione[3]

[1]Department of Soil, Plant and Food Science, University of Bari "Aldo Moro", Via G.Amendola 165/A, I-70126 Bari, Italy; gaetano.balenzano@uniba.it; marialetizia.gargano@uniba.it; [2]Department of Pharmacy-Pharmaceutical Sciences, University of Bari "Aldo Moro", University Campus "Ernesto Quagliarello", via E. Orabona, 4, 70125 Bari, Italy; valeria.ferraro@uniba.it; [3]Department of Agricultural, Food and Forest Sciences, University of Palermo, Viale delle Scienze, B1dg.5, I-90128 Palermo (Italy); fortunato.cirlincione@unipa.it;

According to the principles of the National Recovery and Resilience Plan (PNRR), we focused on the concept of sustainable agriculture and circular economy. The main objective of the project is the eco-sustainable reuse of winery and vineyard waste obtained from the cultivation and processing of "Nero di Troia" CV grapes provided by the company "Trulli il Castagno Snc" Martina Franca (TA), in order to create a growth substrate suitable for the production of mushrooms of food and medicinal interest. The objective is aimed at facilitating the transition of economic activities by promoting the productive reconversion of business activities, the reduction of disposal costs of such waste and pollutant gas emissions. The genus *Pleurotus* (Fr.) P. Kumm. includes economically important mushrooms, cultivated around the world, widely used in human nutrition and recognized as medicinal mushrooms. Within this genus, *P.ostreatus* (Jacq.) P. Kumm., Führer Pilzk. is undoubtedly one of the most widely cultivated and known fungal species in the world. Cultivation was carried out to assess its productive efficiency and qualitative properties on winery and vineyard wastes, in the perspective of a potential large-scale cultivation of wild mushroom species and consequent enhancement of some territories. The cultivation was carried out on five different substrates: wheat straw was identified as the control substrate, pomace, rasps, and pruning clippings each in a 1:1 ratio by dry weight with wheat straw and lastly a mix of the previous substrates in a 1:1:1:1 ratio by dry weight, exhibiting rather good production performance. During the fruiting period, the mycetes exhibited different development depending on the substrate on which they developed. The results of the analysis show that all substrates except the mix showed the same earliness in terms of days. In contrast in fruiting, the mix substrate exhibited a shorter elapsed time from the appearance of primordia to the harvesting of fruiting bodies. In terms of total quantity harvested (grams), the wheat straw substrate showed higher values in contrast to the rasp substrate with the lowest value. The biological efficiency (>50 %) was higher in the wheat straw and pomace substrates than the value (<40 %) obtained in the other substrates. From the analysis of minerals and heavy metals, the rasps showed the highest content [mg/kg(ss)] not exceeding the heavy metal content beyond the legal limits. There are currently other trials underway involving the use of pomace and wheat straw mixed with pomace as a substrate for the growth of *Pleurotus eryngii* (DC.) Quél that may further demonstrate promising results from the perspective of environmental sustainability. Additional analyses on bioactive compounds and biological activities will be carried out, and further trials with other medicinal mushrooms will be conducted.

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Diversity of soil fungi in different beech forest communities of the Wolin National Park (Poland).

Leski Tomasz[1], Rudawska Maria[1], Stasińska Małgorzata[2], Kujawska Marta[1], Karliński Leszek[1], Wilgan Robin[1]

[1]Institute of Dendrology, Polish Academy of Sciences, Parkowa 5, 62-035 Kórnik, Poland, tleski@man.poznan.pl, mariarud@man.poznan.pl, mkowalska@man.poznan.pl, leszekk@man.poznan.pl, rwilgan@man.poznan.pl; [2]Institute of Marine and Environmental Sciences, University of Szczecin, Adama Mickiewicza 16, 70-383 Szczecin, Poland, stasinsk@univ.szczecin.pl

Wolin National Park (WNP) located on the coast of the Baltic Sea is one of 23 national parks in Poland, situated on the Wiolin Island in the north-west part of the country. The continental area of the Park is dominated by moraine hills covered by mixed forests with pine, beech and oak. The main species in the forests is pine, which occupies 68% of the land Park area. Beech covers 23%, oak 7%, and other species the rest of the area. Among forests communities relatively well preserved are beech forests, whose structure is very similar to a natural one. Therefore, our objective was to compare the soil fungal assemblages represented by different functional/trophic groups in four types of beech forest communities: acidophilous beech forest (ABF, *Luzulo-Fagetum*, Natura 2000 habitat type 9110), fertile beech forest (FBF, *Asperulo-*Fagetum, 9130), Pomeranian beech-oak forest (PBOF, *Fago-Quercetum petraeae*), and unique to WNP Baltic orchid beech forest (BOBF, *Cephalanthero-Fagetum*, 9150-5). Soil fungal communities on 20 plots (5 per forest type) were analyzed using high-throughput Illumina MiSeq sequencing of fungal ITS2 amplicons. Among the 632 recorded OTUs Ascomycota and Basidiomycota were the predominant phyla, comprising 61% and 31% of all identified fungi respectivelly. The main functional components of soil fungal assemblages of tested beech forest consisted of ectomycorrhizal (14.7% fungal OTUs), litter saprotrophic (14.7%), soil saprotrophic (11.2%) and wood saprotrophic fungi (10.4%).The highest total OTUs richness was noted in BOBF (299 OTUs) and the lowest in PBOF (266), however, the mean OTUs richness did not differ significantly between different types of beech forests. There were also no differences in the mean OTUs richness for individual trophic groups. Shannon diversity index was significantly different between forest types only for ectomycorrhizal fungi. Only 111 fungal OTUs (17.6%) were shared between all analyzed forest types. The highest number of unshared OTUs was detected in BOBF (173, 27.4%) and the lowest in ABF (32, 5.1%). Analysis of similarities (ANOSIM) and non-parametric manova (NPMANOVA) based on Jaccard and Bray-Curtis dissimilarity indexes revealed that communities of all fungi, as well as individual functional/trophic fungal groups, differ significantly in qualitative (OTUs composition) and qualitative-quantitative (OTUs composition and abundance) structure. Among the identified fungal OTUs, 17 red-listed fungal species were found, comprising ectomycorrhizal, litter saprotrophic, soil saprotrophic, and wood saprotrophic fungi (e.g. *Wynnella silvicola*, *Arrhenia acerosa*, *Entoloma lampropus*, *Boidinia furfuracea*). Our results suggest that the fungal communities displayed high species turnover among forest types rather than differences in species richness, and each analyzed beech forest type helps uphold distinctive fungal diversity.

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How do fungal communities restore in formerly extracted raised bogs.

Martina Vašutová[1], Kamila Vítovcová[1], Karel Prach[1]

[1]Department of Botany, Faculty of Science, University of South Bohemia, Branišovská 31, 37005 České Budějovice, Czech Republic, mvasutova@prf.jcu.cz

Human-made disturbances had and have detrimental effects on peatland ecosystems. The most destroying is the large-scale industrial peat harvesting, accompanied by profound changes in water regime. Possibilities for restoration of such deeply disturbed bogs are limited and their success is mostly evaluated in terms of vegetation and presence of peatland specialist plants. Fungi, although significantly affecting the whole system as decomposers, symbiotrophs or pathogens, have so far received little attention. Therefore, we wanted to find out (1) if succession of fungal communities in disturbed bogs tends to undisturbed bogs and (2) if it follows vegetation succession. Further we were interested (3) if fungal communities associated with a single plant species (*Eriophorum vaginatum*) are affected by bog disturbance. Finally (4) we focused on habitat requirements of six peatland fungal indicator species (*Arrhenia bigelowii, A. gerardiana, A. telmatiaea, Cortinarius chrysolitus, C. davemallochii* and *Galerina hybrida*) to know why they are missing in disturbed bogs. The entire study was conducted in the Šumava National Park (the Czech Republic) and the surrounding area. Fungal communities were studied in detail in 24 permanent plots (9 x 9 m) in two disturbed bogs with the occurrence of various successional stages up to the 35 years, 8 plots were established in two closest undisturbed bogs as a reference. Fruit body survey (2019-2020, 6 times per season) and Illumina sequencing of *Sphagnum* and peat detected different parts of fungal communities. Fruit bodies data reflected vegetation pattern better than sequence data because they included many specialized saprotrophs living on dead plant litter or wood. In agreement with a successional trend in vegetation, fungal communities of old stages did not resemble these of undisturbed bogs. Altogether 30 *E. vaginatum* tufts in disturbed bogs and 20 tufts in undisturbed bogs were analysed using Illumina sequencing. Disturbance had a significant effect on communities in both, living and dead leaves of *E. vaginatum*. Based on the analysis of the vegetation around 144 collections of fruitbodies of peatland fungal indicator species from 61 peatlands we found out that they preferred sites where peatland specialist plants occur. To summarize, fungal communities follow vegetation succession, similarly, they do not move towards undisturbed bogs. Moreover, as seen in *E. vaginatum*, even when the right plants are present, associated fungal communities can vary. Studied fungal indicator species are probably unable to establish on disturbed bogs from the similar reasons as peatland specialist plants which include low water table, low remaining peat and possibly dispersal limitations. Information obtained from Illumina sequencing of fungi is still difficult to interpret because many unknown operational taxonomic units were detected in all datasets.

eDNA metabarcoding data in GBIF.org.

Dmitry Schigel

Global Biodiversity Information Facility

The Global Biodiversity Information Facility (GBIF) is an international network and data infrastructure that provides free and open access to biodiversity data from around the world, enabling scientists, policymakers, and the public to explore and analyze information about the Earth's living organisms. Metabarcoding of DNA is becoming a standard tool for detecting species in bulk samples or environmental samples such as soil, water, and air. In the name of reproducibility, raw sequence data (fastq files), code and metadata files are most often shared and deposited in dedicated repositories within the International Nucleotide Sequence Database Collaboration, GitHub, and in generalist repositories such as Dryad and Zenodo. However, seen from a biodiversity perspective, raw sequences have narrower applications compared to the interpreted DNA metabarcoding data. Most often such interpretations come in the shape a table with sequence abundances of Amplicon Sequence Variants or Operational Taxonomic Units across samples – a so-called ASV/OTU-table – and some associated files, e.g. spatiotemporal and other sample metadata, and taxonomic inferences of sequences. In most cases these are at best part of the supplementary materials, and unfortunately rarely shared in a standardized way that facilitates reuse and integration of DNA data with other data types and data layers, and across studies. In this session, we present GBIF plans and developments on improving mediation and standardisation of OTU-table like biodiversity data for easier and broader reuse, and integration with other biodiversity data and data layers, with reference to a recent survey in the GBIF network.

Keywords
Environmental DNA, OTU tables, FAIR data, Biodiversity

Effect of ecosystem changes on proliferation of tow ecotypes of Morels (*Morchella* spp.).

Segula Masaphy

Applied Mycology and Microbiology Dept., MIGAL – Galilee Research Institute, PO Box 831, Kiryat Shmona 11011, and Tel Hai College, Upper Galilee 12210, Israel

An increasing understanding of the impact of environmental factors on mushroom temporal and spatial proliferation in nature is important for conservation and control of mushroom populations, especially but not exclusively, with regard to edible mushrooms. Both local climate changes and site-related environmental changes such as disruptive activities, may affect mushroom proliferation. Morels (*Morchella* spp., Pezizales) are important edible mushrooms known for their delicate taste and aroma, as well as their medicinal properties. Morchella species exhibit a wide range of trophic modes, being mycorrhizal, endophytic or saprotrophic in nature. Morels usually appear for only a short duration in a season and in wide range of habitats. The proliferation pattern of two distinguished Morchella species, exhibiting mycorrhizal and free-living saprotrophic modes, was monitored in Israel together with climatic and soil condition data. Results: air climatic changes were found to affect the temporal fruiting pattern of morels as well as the mushroom phenotypic and chemical composition properties such as color and phenolic contant. Moreover, both population types were affected by disruptive activities such as fires and site forest management activities.

Emerging pathogenic fungi. Emergence of the wood-decay pathogen *Fomitiporia mediterranea* in Pavia province (North Italy).

Carolina Elena Girometta[1], Rebecca Michela Baiguera[1], Anthea Desiderio[1], Simone Buratti[1], Silvano Lodetti[1], Francesco Bracco[1], Elena Savino[1]

1Department of Earth and Environmental Sciences, via Sant'Epifanio 14, 27100 Pavia, Italy,
carolinaelena.girometta@unipv.it

Since *Fomitiporia mediterranea* has been recognized to be an independent species in 2002, it has been increasingly related to wood decay diseases and especially esca disease in grapevine. Moreover, its genome has been completely sequenced and currently represents a model for white rot agents.

Pavia province includes two main macroareas: lowlands and Oltrepò Apennines. The former are intensely cultivated except for limited areas close to Ticino and Po rivers, whereas the latter includes an intensely cultivated belt (mainly vineyards and herbaceous crops) and woodland/meadows in the highest belt.

Since 2010 the DSTA-Unipv Laboratory of Micology has mapped the presence and ecological preferences of wood decay species in several areas. Moreover, a major task of our Laboratory is to achieve strains in pure culture, that means we have got the molecular confirmation of identification for a significant subsample in order to exclude possible specimens of the sister species *F. punctata* (Girometta et al., 2020; Cartabia et al., 2022).

Aim of the present contribute is to report the frequency and environmental preferences of *F. mediterranea* in Pavia province based on a 13 years-monitoring and mapping.

Based on our data, *F. mediterranea* has increased its presence in both the lowlands and Apennine hills, whereas is still apparently excluded from the mountain belt. This species is most widely represented on *Vitis vinifera* (vineyards) and *Robinia pseudoacacia*, although its host spectrum also includes *Corylus avellana*, *Olea europaea* and *Punica granatum*.

Our visual field mapping highlights that basidiome development begins on dead wood (logs, stumps, standing trees) or living trees depending depending on the host and suggesting different behaviours. In fact, basidiomes always occur on dead *V. vinifera* and dead *C. avellana* sticks, whereas they can appear on declining but still living *O. europaea*, *P. granatum* and *R. pseudoacacia*. It should be highlighted that *F. mediterranea* is also frequent on dead *R. pseudoacacia*.

These results therefore suggest *F. mediterranea* behavior is mostly necrotrophic in *V. vinifera* and *R. pseudoacacia*, whereas it shifts towards weak pathogenicity or saprotrophism in other hosts. Increasing occurrence of basidiome records on dead hosts suggest this species is an emerging pathogen in vineyards (Apennine hills) and especially in *R. pseudoacacia* (both Apennine hills and lowlands). On the other hand, *C. avellana* in lowlands woods is apparently affected by saprotrophism more than necrotrophism.

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Emerging ink disease of sweet chestnut in southern Switzerland.

S. Prospero[1], M. Heinz[1], J. Engelbrecht[2], P. Fonti[1]

[1]Swiss Federal Institute for Forest, Snow and Landscape Research WSL, Birmensdorf, Switzerland, [2]Department of Microbiology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa

In southern Switzerland, sweet chestnut (*Castanea sativa*) is the dominant tree species up to about 900 m a.s.l., where it forms a continuous forest belt of about 30’000 hectares. Since the 1990s, an increasing mortality of chestnut trees has been observed. The specific symptoms shown by the declining trees (i.e., thinning of the crown, small chlorotic leaves, necrotic lesions at the root collar) suggested an emergence of ink disease. In this study, we determined 1) the current distribution and the causal agents of ink disease, 2) the population structure of *Phytophthora cinnamomi*, 3) the dynamic of an infection by *P. cinnamomi* and *P. x cambivora*, and 4) which tree species could potentially replace sweet chestnut in infected sites. We identified a total of 25 outbreaks, 19 caused by *P. cinnamomi*, 5 by *P. x cambivora*, and one by both species. Dendrochronological analyses revealed that the disease emerged in the last 20–30 years. Infected trees either die rapidly within 5–15 years after infection or show a prolonged state of decline until death. The genetic structure of the *P. cinnamomi* population suggests both independent introductions and local spread of the pathogen. A modeling approach showed that native species like Norway maple (*Acer platanoides*), sycamore maple (*Acer pseudoplatanus*), large-leaved lime (*Tilia platyphyllos*), European hop-hornbeam (*Ostrya carpinifolia*) and common whitebeam (*Sorbus aria*) could potentially replace sweet chestnut under two scenarios of climate change.

Endophytic and soil fungi in ecosystems polluted with toxic metals.

Rafał Ważny[1*], Magdalena Zyzik[1,4], Roman J. Jędrzejczyk[1], Agnieszka Domka[2], Artur Pliszko[3],
Weronika Kosowicz[1,4], Maciej Gustab[1,4], Piotr Rozpądek[1]

[1]Małopolska Centre of Biotechnology, Jagiellonian University in Kraków, Gronostajowa 7a, 30-387 Kraków, Poland,
[2]W. Szafer Institute of Botany Polish Academy of Sciences, Lubicz 46, 31-512 Kraków, Poland
[3]Institute of Botany, Jagiellonian University in Kraków, Gronostajowa 3, 30-387 Kraków Poland
[4]Doctoral School of Exact and Natural Sciences, Jagiellonian University in Kraków, S. Łojasiewicza 11, 30-387 Kraków, Poland
*rafal.wazny@uj.edu.pl

Symbiotic fungi play a key role in plant adaptation to the environment. Very little is known about how metal toxicity impacts the plant microbiota and about the relationship between plant and soil microbiota under metal toxicity. The aim of this study was to evaluate the effect of toxic metals present in the soil on the biodiversity endophytic mycobiota inhabiting plant seeds, shoots and roots.

The mycobiome of model plant *Arabidopis arenosa* and soil from a Zn-Pb heaps and metal-free ruderal sites were compared. The biodiversity of fungi inhabiting mine dump substrate was lower than that of the metal free site. Metal toxicity limited *Agaricomycetes* and *Leotiomycetes* in the soil. In the leaf and root endosphere of *A. arenosa* from the mine dump the number of endophytic fungal taxa was comparable to that in the reference population, but the community structure significantly differed. *Agaricomycetes* was the most notably limited class of fungi in the leaves and roots. This was verified in a reconstitution experiment in the laboratory.

In the case of seeds we did not observe any effect of soil toxicity on the alpha and beta diversity of endophytic fungi inhabiting plant seeds. The results indicate that seed mycobiome is constitutive and resistant to the toxicity of heavy metals and that vertical transmission shapes seed mycobiome more than horizontal transmission. This hypothesis is verified in experiment focused on vertical transmission of endophytes in consecutive plant generations. Endophytes present in the seeds of plants exposed to toxic metals might help to survive a new plant generation and spread out into rhizosphere and surrounding soil, shaping the biota of environments that are at the starting point of succession.

The results presented in our study indicate that metal toxicity affects the structure of the plant mycobiota not by changing the pool of microorganisms available in the soil from which the fungal symbionts are recruited but most likely by altering plant and fungi behaviour and the organisms' preferences towards associating in symbiotic relationships. It is worth to be studied if this phenomenon is universal for other plant species inhabiting polluted sites.

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Environmental factors affecting content of nutrients in mushrooms.

Katarzyna Stojek[1], Barbara Bobrowska-Korczak[2], Bogdan Jaroszewicz[3]

[1]University of Warsaw, Białowieża Geobotanical Station, Sportowa 19, 17-230 Białowieża, Poland, km.stojek@uw.edu.pl; [2]Medical University of Warsaw, Faculty of Pharmacy, Stefana Banacha 1, 02-097 Warszawa, Poland, barbara.bobrowska@wum.edu.pl; [3]University of Warsaw, Białowieża Geobotanical Station, Sportowa 19, 17-230 Białowieża, Poland, b.jaroszewicz@uw.edu.pl;

Mushroom picking is a popular recreational activity in many European countries. The areas where people collect mushrooms are often carefully selected to maximize the harvest size and the probability of encountering the most flavorous species. However, the nutritional value is rarely considered and often underestimated, even if mushrooms can be an essential diet element, especially for diabetes and vegans. Mushrooms are characterized by rich amino acid composition, remarkable vitamin content, and low content of fats, but with a high share of polyunsaturated fatty acids, that are essential for our health[1,2].

We investigated the environmental factors, with particular emphasis on stand characteristics (tree species identity and richness), affecting mushroom productivity and nutritional values. Our study was conducted at sites in Poland, Belgium, and Germany, in plots located under similar environmental conditions (i.e., topography, geology, soil type) but varying in tree species richness and composition. We present our results on the impact of environmental factors on productivity and nutritional values (fatty acids, proteins) of mushrooms. The composition of FA was analysed using GC-MS (gas chromatography – mass spectrometry), whereas the proteins content by Kjeldahl method. To estimate the impact of environmental factors we used mixed-linear modelling, PCA and heatmaps.

We revealed that fatty acids (FA) content and composition in mushrooms was mainly affected by fungus species (explaining 27-33% of the total variation). Intraspecies differences in fatty acids (FA) content were partly explained by environmental factors. Increasing C/N ratio and decreasing N concentration affected positively the total content of FA, unsaturated FA, and saturated FA. Canopy and understory cover affected composition of FA. Tree species richness and composition did not influence the composition of FA, except for single species, *Rhodocollybia butyracea*, collected in the German site, which differed between monocultures and mixed stands.

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Environmental study on different *Tuber magnatum* natural production areas.

Mara Rondolini[1], Gilberto Bragato[2], Leonardo Baciarelli Falini[1], Domizia Donnini[1]

[1]Department of Agricultural, Food and Environmental Sciences, University of Perugia, Borgo XX Giugno 74, 06121 Perugia, Italy; [2]CREA VE - Research Centre for the Soil–Plant System, Viticolture Via Trieste 23, 34170 Gorizia, Italy

The white truffle (*Tuber magnatum* Picco) is an hypogeous ascomycete belonging to the *Pezizales* order and *Tuberaceae* family. The ascomata of *T. magnatum* is one of the most valuable food products for its particular aroma, which is a result of its association with other microorganisms in addition to environmental factors (1,2). Although other truffle species are widely cultivated, the production of white truffles is still derived from wild harvesting. However, the areas where *T. magnatum* is developing, are involved in a significant decrease due to climatic change, lack of maintenance, and overexploitation of the resource. In that sense, finding ecological characteristics related to white truffle presence becomes essential to preserve these environments and it assumed an unestimated economic and ecological value. This study aims to describe two different *T. magnatum* natural production areas to find which are the key aspects of its development. In particular study areas are located in the center of Italy: one in Umbria and one in Tuscany. The latter corresponds to the typical valley floor environment, located among the particular landscapes calls "crete senesi" of Tuscany's south. However, the other one in Umbria is an along river site, hence is correlated to all riparian environmental conditions. For each area were collected soil, vegetation, and climatic data. The purpose is to find which are the correlations between such different environments with the presence of white truffles due to clarify some ecological aspects are still unclear.

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Ethanol extracts of *Fomes inzengae* and *Laetiporus sulphureus* inhibits in vitro survival and migratory activity in hepatocarcinoma cells.

Lara Lizzi, Martina De Mattheis, Federica Russo, Marco Leonardi, Mirco Iotti, Mara Massimi

Department of Life, Health & Environmental Sciences, University of L'Aquila, Coppito 1, via Vetoio, 67100 L'Aquila, Italy

Fomes inzengae (a sister species of *F. fomentarius*) and *Laetiporus sulphureus* are lignicolous species, parasite of deciduous trees and widely distributed in Europe. These species like other members in the same genera are a source of many bioactive compounds such as polysaccharides, triterpenes and their derivatives, lipids, and secondary metabolites. These substances are studied for their anticancer, immunomodulatory, anti-inflammatory, and antiviral properties. Antitumoral activity of *F. fomentarius* extracts was studied on different cell lines but not in hepatocarcinoma cells. *L. sulphureus* demonstrated antiproliferative effect on HepG2 cells, although the molecular mechanisms involved in antitumoral activities have not been investigated. We investigated the in *vitro* effects of ethanol extracts of *F. inzengae* and *L. sulphureus* on two hepatocarcinoma cell lines, HepG2 and Huh7. To determine the molecular mechanisms underlying anti-cancer effects, various methods were used, including cell counting (Trypan Blue Exclusion Test), DAPI staining, cytotoxicity tests and Western Blot. *F. inzengae* and *L. sulphureus* extracts significantly inhibited proliferation of the HepG2 cell line in a time and dose-dependent manner. Treated cells showed a lower vitality and formed smaller aggregates. The significant increase in the protein expression levels of the tumor suppressors p53, p21 and p27, suggested an inhibition in the G_1 cell cycle phase. In addition, our results showed a reduction of the anti-apoptotic protein Bcl-2 and increased levels of the pro-apoptotic proteins Bax, as well as of cleaved caspase-9 and -3. We also found that extracts inhibited Huh7 migratory activity, an important event involved in cancer invasion and metastasis, characterized by downregulation of E-cadherin, β-catenin and upregulation of Twist, Snail, Slug. The results of Western blot experiments confirmed the inhibition of Huh7 invasion with a significant increase in E-cadherin expression and reduction in Twist protein levels.

Although further studies are needed to investigate and clarify the anticancer potential of these polypores, we believe they could be useful to support anticancer therapies.

Evaluation of global diversity of leaf-associated fungi.

Niloufar Hagh Doust[1,2], Leho Tedersoo[1,2]

[1]Institute of Ecology and Earth Sciences, Univerisy of Tartu, Tartu, Estonia, E-mail Niloufar.hagh.doust@ut.ee
[2]Mycology and microbiology center, University of Tartu, Estonia

Plant individuals function as holobionts, providing a variety of microhabitats that accommodate diverse microbial lineages, both endophytic and epiphytic. These microorganisms colonize the belowground and aboveground surfaces of plants, as well as their internal tissues. The interactions between plants and their microbiomes play a crucial role in ecosystem functionality. The aboveground parts of plants serve as hosts to a multitude of ecologically significant fungi and bacteria, encompassing pathogens, endophytes, and epiphytes. Despite their importance, the distribution patterns and diversity of these microorganisms remain enigmatic. Our project's primary aim is to unravel the global diversity of bacteria and fungi associated with leaves while investigating the key factors that shape their communities. In this overview, I will discuss the ongoing project, including our hypotheses and various aspects of the research. Furthermore, I will delve into the methods employed, the challenges encountered, and the progress achieved thus far.

Exploiting wood-destroying fungi for the fabrication of novel biomaterials: mycelium composites.

Ilze Irbe[1], Inese Filipova[1], Laura Andze[1], Marija Tereze Dzierkale[2,1], Marite Skute[1]

[1]Latvian State Institute of Wood Chemistry (LS IWC), Dzerbenes iela 27, Riga, LV-1006, Latvia, ilze.irbe@kki.lv;
[2]University of Latvia, Raina bulvaris 19, Riga, LV-1586, Latvia, marijadzierkale@inbox.lv

Wood-destroying fungi play a significant role in the natural decomposition of wood in forest ecosystems. However, recent research has explored the potential of these fungi in the fabrication of novel biomaterials, particularly mycelium composites (MC). When combined with lignocellulosic substrates such as agricultural waste, sawdust, or others, mycelium can be utilized as a versatile biofabrication tool to create innovative biomaterials. This process, known as mycelium-based self-assembly, offers a sustainable alternative to traditional manufacturing methods (Fig. 1).

Fig. 1. Fabricated mycelium composites

This study provides an overview of MC created by the Cellulose laboratory (LS IWC) team. Two different types of agricultural and forestry waste, namely hemp shives and birch sawdust, as lignocellulose substrates were used. These substrates were bound together by the natural growth of mycelium of basidiomycete *Trametes versicolor*. The physical, mechanical, water-related properties, and biodegradability were determined. The key findings indicate that the growth of mycelium resulted in partial degradation of the lignocellulosic substrates, with cellulose content reduced by 7% to 28% and lignin by up to 13% from their initial levels. The mechanical properties, specifically compression strength (CS) and bending strength (BS), were influenced by the density of the samples. A larger granulometric fraction of hemp MC ensured lower density and consequently higher mechanical strength, but weakened water absorption performance. On average, hemp MC achieved CS values of 0.18 MPa and BS values of 0.12 MPa, while sawdust MC exhibited CS and BS values of 0.20 MPa and 0.21 MPa, respectively. Water absorption was high, ranging from 350% to 850%, with lower values observed for sawdust MC. Nevertheless, the volumetric swelling of the MC samples ranged only from 2% to 8%, which was lower than that of traditional composites like particle board, plywood, or OSB. The hygroscopic sorption isotherms demonstrated promising results, with only a slight moisture increase up to 14% in an air environment with 85% relative humidity. The thermal conductivity (λ) for hemp MC was approximately 40 mW/mK, while for sawdust MC types, it was around 48 mW/mK. Biodegradation by the composting method demonstrated that all MC specimens completely disintegrated after 12 weeks of exposure to natural soil microbiota. The MC exhibited acceptable mechanical properties, including high strength-to-weight ratios and good insulation properties, making them competitive to some commercial expanded polystyrene (EPS) types. The MC materials have potential applications in various fields, such as architecture, product design, and packaging. They can be used as insulation panels, furniture, packaging materials, and as biodegradable alternatives to traditional plastics.

Exploring Macrofungal Diversity in Sacred Natural Sites of Umbria, Italy.

Giancarlo Angeles Flores[1], Gaia Cusumano[2], Federica Pietrelli[2], Ilaria Proietti[2], Roberto Venanzoni[2], Paola Angelini[2]

[1] Botanic Garden “Giardino dei Semplici”, Department of Pharmacy, “Gabriele d’Annunzio” University, 66100 Chieti, Italy; giancarlo.angelesflores@unich.it.
[2] Department of Chemistry, Biology and Biotechnology, University of Perugia, 06122 Perugia, Italy; gaia.cusumano@studenti.unipg.it, federica.pietrelli@studenti.unipg.it, ilaria.proietti@studenti.unipg.it, roberto.venanzoni@unipg.it, paola.angelini@unipg.it

Over the past decade, an increasing number of studies have highlighted the crucial role of Sacred Natural Sites (SNSs) in nature conservation. SNSs represent another instance of nature preservation that operates separately from Protected Areas, even though they frequently share the same geographic space. SNSs are defined as “areas of land or water that hold special spiritual significance for peoples and communities”. These sites are associated with various natural features, including individual trees, rivers, mountains, islands, and even entire landscapes, with the majority being sacred groves and forest patches. Occasionally, SNSs may also incorporate built structures like temples, shrines, or monasteries, surrounded by natural or semi-natural areas. The primary objective of this study is to explore the macrofungal diversity and community assemblages in three sacred groves dominated by *Quercus ilex* L. in the region of Umbria. Over a period of 1 year, 492 basidiomata and ascomata were collected from the SNS, revealing a total of 132 taxa from 37 families and 73 genera. The distribution of relative abundance was analyzed using a rank–abundance (dominance–diversity) curve. Macrofungal species diversity was determined using Fisher’s alpha (F), Shannon (H), and Simpson (1/D) indices (Angelini et al. 2016). The study also examined the similarity of macrofungal communities among the three sacred groves using similarity indices based on presence/absence data (Jaccard's index) and relative abundance (Morisita–Horn index) (Angelini et al. 2017). The “Lecceta of Sassovivo” (SCI IT5210042) displayed higher mycorrhizal species richness, while the “Monteluco of Spoleto” (SCI IT5210064) and “Ditch of the Eremo delle Carceri” (SCI IT210030) had a higher relative number of saprotrophic macrofungi. The macrofungal community of *Q. ilex* woodland in “Monteluco of Spoleto” exhibited greater richness and diversity compared to the woodland types of the other sites. Despite the need for further sampling to compile a comprehensive macrofungal inventory, the 2022 survey revealed that all of the sites studied support a habitat for various macrofungal species, including ectomycorrhizal species.

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Exploring the Fungal Biodiversity Richness in the Root System of Lavender.

Vittorio Capra [1], Laura Canonica [1], Grazia Cecchi[1], Simone di Piazza[1], Mirca Zotti[1]

[1]DISTAV - University of Genoa, Corso Europa 26, Italy, vittorio.capra@edu.unige.it, laura.canonica@edu.unige.it, grazia.cecchi@edu.unige.it, simone.dipiazza@unige.it, mirca.zotti@unige.it

Since the beginning of microbial communities' investigations, a plethora of novel discoveries has taken place in science, from the medical to the commercial fields. In particular, soil studies have linked many of the different living kingdoms sharing a plant roots' space in a tapestry woven as a mix of cooperation and competition (1). Few very known examples (like Nitrogen-fixating bacteria and mycorrhizal fungi) have been enriched with new lists of microscopic prokaryotes and eukaryotes favoring root growth, leaf size, nutrient absorption, photosynthetic capability, or pathogen resistance (2). To address how and which fungal organisms influence plant physiology and can be better employed to improve plant properties, an in-depth characterization of culturable fungal isolates from Lavender's root system has been accomplished. Lavender plants were chosen because of their historical use in many modern products, from cosmetics to hygiene and food production (3). This has led to a solid framework of quantitative traits and properties to be checked in order to study the effects on plants of potential treatments, such as a planned modification of the fungal mycorrhizal community to strengthen the presence of Plant-Growth-Promoting-Fungi (PGPF). Soils from Lavender fields were sampled and its culturable fungal component morphologically and molecularly characterized. Sampling sites included both Mediterranean and Alpine climates, respectively the Albenga plains and Colle di Nava. Agricultural practices, soil acidity and constitution, the presence or absence of Lavender spontaneous growth have all been taken into consideration when comparing the mycorrhizal community composition, in conjunction with biodiversity indexes computation. As a result, isolated species belonging to sixteen different genera have been identified and described, and novel species discovered. Interestingly, fungi from the more thermophilic *Aspergillus* genre could be found only close to the Mediterranean Sea (Albenga plains), while the novel species localization differed as well among the sampling sites. As a conclusion, plant growth promotion potential was examined via Solubilization Index (SI) measurement in both CAS agar and Pikovskaya agar plates, common tests used to check for siderophore production and phosphate solubilization, respectively. The present study constitutes an introductory work pipeline linking soil and microbiota characterization to both taxonomic advances and testing for in-the-field applications.

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First evidence of symbiotic relations between *Bradyrhizobium* spp. and *T. magnatum* mycelium.

Simone Graziosi[1], Federico Puliga[1], Mirco Iotti[2], Alessandra Zambonelli[1]

[1]Department of Agricultural and Food Sciences, University of Bologna, Viale G. Fanin 44, 40127, Bologna, Italy, simone.graziosi5@unibo.it; federico.puliga2@unibo.it; alessandr.zambonelli@unibo.it; [2]Department of Life, Health and Environmental Science, University of L'Aquila, Via Vetoio, 67100, Coppito, L'Aquila, Italy, mirco.iotti@univaq.it

The mycelium of *T. magnatum* Picco has never been grown reliably in vitro conditions until now. *T. magnatum* ascoma hosts a specific bacterial community generally dominated by Alphaproteobacteria with *Bradyrhizobium* spp. the most abundant taxa. Several studies hypothesized that these bacteria may have a specific role on the formation, nutrition and maturation of ascomata but no information are reported on their possible role on *T. magnatum* mycelium development. The present study aimed to describe the effects of *Bradyrhizobium* spp. on isolation and development of *T. magnatum* mycelium. After many attempts, three *T. magnatum* strains were co-isolated with bacteria on mWPM (Iotti et al., 2005) from the gleba of immature ascomata. Specific tests showed that both the isolated bacteria and *T. magnatum* mycelia were unable to grow alone in mWPM. Moreover, the isolated bacteria did not positively affect the growth of mycelia of other truffle species (*T. borchii*, *T. melanosporum*). Phylogenetic analyses inferred by *16S*, *glnII* and *recA* genes showed that isolated bacteria belong to the *Bradyrhizobium* genus and that they are grouped in a clade close to *Bradyrhizobium elkanii. nif*H genes were detected in all the bacterial isolates. Our results suggest a specific obligate symbiosis between these Bradyrhizobia and *T. magnatum* mycelium. Although further studies are necessary to better understand the mechanisms of interaction between *T. magnatum* and *Bradyrhizobium* spp., these results could have important biotechnological applications in truffle cultivation.

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From data mining to data meaning in fungal evolution of diversity.

Gianluigi Cardinali

Department of Pharmaceutical Sciences – University of Perugia

Fungi are a phylum of exceptional complexity due to a long evolutionary history and to an extraordinary genetic plasticity. The possibility to have both sexual and asexual reproduction or to switch between these two forms, the active horizontal gene transfer and an unsuspected frequency of interspecific hybridizations give fungi a full set of mutational and recombination tools. All these aspects have produced the current biodiversity of this phylum, that we know only from cultivated and culturable species.

The vast arsenal of cultural, metagenomic and metabolomic techniques to unveil the evolutionary of Fungi, produced the increasingly popular concept is that, once data are available, their meaning will magically pop-out from the appropriate strategy of data mining. This paradigm tends to overcome the hypothesis driven approach that has been the backbone of scientific development from the early time of the scientific method.

These epistemological aspects pose several questions on the optima strategy to follow for the analysis of the increasingly larger amount of data accumulating in public databases.

The presentation will compare on a conceptual basis the data mining vs the hypothesis driven system and the possibility to make the two approaches cooperating for a deeper understanding of fungal biology and evolution. Data mining require infrastructures, advanced algorithm development, availability of good quality data and increasing introduction of IT and bioinformaticians in biology. On the other hand, the application of the hypothesis driven approach requires a quest for reliable models to develop hypotheses and experimental designs. In both cases, there is increasing evidence that biologists need to cooperate in a transdisciplinary manner with other fields of science and even of philosophy. The concept and the praxis of species delimitation are at the core of the understanding of the actual biodiversity distribution and of its evolutionary development. This aspect as well as case studies based on datasets of mycological interest will be presented and discussed.

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Fungi in changing ecosystems.

Simone Di Piazza[1]

[1] Dipartimento di Scienze della Terra, dell'Ambiente e della Vita, Università degli Studi di Genova,Corso Europa 26, 16132 Genova, Italy; mirca.zotti@unige.it

Changes in ecosystems can be caused by natural or human induced extreme events. In both cases the change can be sudden (e.g. floods, fires, drought, oil spill) or more or less gradual (e.g. diseases, industrial pollution, deforestation, light pollution). Today ecosystem changes are more and more frequent and, often directly or indirectly, connected to human activities. In particular, humans have caused deforestation, introduced invasive species, induced soil, water and air pollution: all of which affect ecosystems health more or less severely and require external interventions to mitigate the damages. In this context our question is, what could be the role of fungi in these changes? Fungi are an important component in the majority of Earth's ecosystems. They are very heterogeneous organisms with great diversity in function, reproduction, biological and non-biological interactions. Thanks to their great diversity, fungi are able to colonize a wide range of environments mediating the relationship among different organisms and substrates. Therefore, the mycobiota help buffer these changes by acting as a driving ecological force to preserve ecosystems health and functions. As further evidence of the great potential of fungi in this context, there is an extensive scientific literature from researchers spread all around the world.

This presentation is devoted to describing the main projects carried out by the group of University of Genoa in the last ten years. The results achieved prove that fungi can be fruitfully exploited to remediate/mitigate ecosystems pollution. Moreover, some case studies will be also presented where fungi can have a different role (harmful or threatened) stimulating the discussion about the positive and negative importance of fungi in ecosystem balances.

Fungal Data Management & Open Science. Developing a Network for Macromycetes Biodiversity: A Case Study from Italy.

Speakers: Francesca Floccia[1], Edoardo Velli[2],

[1]Italian Institute for Environmental Protection and Research, Rome, Italy;
[2]Italian Institute for Environmental Protection and Research, Ozzano dell'Emilia (BO), Italy

In Italy, the study of mycological diversity has been hindered by the lack of specific legislation pertaining to the monitoring and conservation of fungal species, resulting in limited attention from national institutions. However, valuable knowledge exists at the local scale, albeit geographically scattered and unorganized, impeding its full utilization and interoperability.

To address this challenge, in 2021, the Italian Institute for Environmental Protection and Research (ISPRA) started the Network for the study of mycological diversity, with the engagement of mycologists as "citizen scientists" in a voluntary, cost-free, and autonomous initiative. The primary objective of the Network is both gathering new data on fungal biodiversity in specific habitats and harmonizing existing data at a national level, thereby providing established methodologies, standards, and guidelines, as well as web and mobile tools, ensuring the collection of Findable, Accessible, Interoperable, and Reusable (FAIR) data.

The project’s data management process comprises three main stages: data collection using freely available web and mobile applications, electronic archiving and organization of data, and translation into accessible information for citizens. To ensure control and repeatability, the entire process underwent ISO 9001:2015 certification.

Additionally, after one year, the Network expanded its efforts to include the collection of biological samples. Consequently, a national *fungarium* was established, enabling the conservation of fungi specimens, and facilitating genetic analysis to aid in taxonomic determinations.

After two years of activity, the Network has successfully developed a national database and engaged mycologists in its initiative. Genetic protocols for DNA extraction and sequencing, as well as innovative analytic approaches have been tested and applied to the first fungal samples.

In conclusion, we present an experimental data management process that addresses the challenges associated with harmonizing mycological biodiversity data. Our ongoing focus is to disentangle systematic and taxonomic ambiguities in classifying fungal taxa, enhancing our understanding of the intricate world of fungi.

Fungi associated with ambrosia beetles in southern beech forests of Patagonia, Argentina.

Williams G. [1], Molina L. [2,3], de Errasti A.[2,3], Pildain M. B. [2,3,4]

[1] USDA Forest Service, USA
[2] Centro de Investigación y Extensión Forestal Andino Patagónico (CIEFAP), Argentina
[3] Comisión Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina
[4] Universidad Nacional de la Patagonia San Juan Bosco, Argentina
mbpildain@ciefap.org.ar

In temperate forests, bark and mycetophagous ambrosia beetles rely on, directly interact with, and help to shape fungal communities in addition to playing important roles in stand turnover and as landscape disturbance agents. Worldwide, fungal symbionts have been studied in fewer than 5% of described ambrosia beetle species. In *Nothofagus* (Southern beech) forests of Northern Patagonia, there have been many studies on mortality and decay involving fungal agents, but fewer studies of ambrosia beetles or of their fungal associations. We sampled galleries, adults and larvae of *Gnathotrupes* (Curculionidae, Coleptera) in monospecific stands of lenga (*N. pumilio*) across four sites affected by mortality and decay within Los Alerces National Park (PNLA) in Chubut Province, Argentina. In each site, five trees with evidence of recent wood-borer activity were sampled. In five galleries per tree, larvae or adults were collected, isolations were made from the galleries and the bodies of the insects and fungi were identified by morphological and molecular techniques (ITS region). A total of 95 isolates revealed a taxonomically and functionally diverse assemblage of fungi including plant pathogens, saprotrophs, and entomopathogens. The most common fungi from plant pathogenic genera were blue stain fungi, *Ophiostoma patagonicum* and *Leptographium gestamen*, followed *Ilyonectria* and *Huntiella decorticans*. The most common saprophytic fungi were the yeast *Cyberlindnera*. Wood decay-causing Basidiomycota and species of the ambrosial genus *Raffaelea* were also present. The only entomopathogen that was isolated belonged to *Beauveria*, and could play a role regulating beetle populations. The results demonstrate diversity across fungal functional guilds, including fungi that play roles in nutrient cycling and decline in tree health, potential options for biological control of insects, and the first ambrosial fungi described from *Gnathotrupes* spp.

Fungi sniffing fungi: a working expansion of the Phenotype MicroArrays™ system to assess the effects of fungal volatile organic compounds between co-occurring species.

Veronica Spinelli[1], Andrea Ceci[1], Flavia Pinzari[2] and Anna Maria Persiani[1]

[1]Department of Environmental Biology, Sapienza University of Rome, Piazzale Aldo Moro 5, Rome, Italy, veronica.spinelli@uniroma1.it, andrea.ceci@uniroma1.it, annamaria.persiani@uniroma1.it ; [2] Institute for Biological Systems, National Research Council of Italy (CNR), Area Territoriale di Ricerca RM1 Strada provinciale 35d, 9, Montelibretti, Rome, Italy, flavia.pinzari@cnr.it

Fungal volatile organic compounds (FVOCs) have been reported to act as intra- and inter-kingdom communication signals. FVOCs, considered long-distance messengers, can influence, either positively or negatively, the growth and/or behaviours of other soil organisms involved in relationships with fungi (1,2). In fact, it has been demonstrated that fungal VOCs can interfere with essential metabolic pathways to prevent fungal growth, and determine a growth stimulation (2). From an ecological perspective, fungal VOCs, representing important carbon sources, can be crucial for pioneer fungi colonizing carbon-limited niches. Instead, in less limiting conditions, several fungal VOCs, acting in a concentration-dependent way, can regulate mycelium growth and spore germination in their environment and participate in reciprocal cross-talk. So as developmental signals, they acquire particular importance during population establishment (2). Hence, gaining insights into the semiochemical aspects of interactions between co-occurring species that occupy similar niches is of great significance for fungal ecology (1). The Biolog® Phenotype MicroArrays™ (PM) system allows studying phenotypic profiles and metabolic responses of fungi on many different carbon sources. PM system is a powerful tool for studying niche overlap, the level of ecological similarity between single species and the catabolic versatility of saprotrophic fungi (3). Understanding the effect of the VOCs of one fungal species on the metabolism of another species and evaluating niche modification could also represent a valuable assessment to better calibrate the formulation of synthetic consortia of soil strains for biotechnological applications (4). Therefore, in this study, we propose a working expansion of the Biolog® Phenotype MicroArrays™ (PM) system for assessing the effect of the VOCs of one fungal species on the metabolic response of another species. The proposed system has been developed to study the interactions between two strains of soil saprotrophic fungi currently preserved at the culture collection of the Fungal Biodiversity Laboratory (FBL) (Sapienza, University of Rome): *Minimedusa polyspora* FBL 503 and *Chaetomium globosum* FBL 205. The proposed VOC-PM system experimental configuration was as reported in figure 1. A 96 well plate, after four days of fungal growth on potato dextrose agar, was covered with a sterile sealing film having a 0.80 mm diameter hole in the centre of each well, allowing VOCs diffusion, and was placed upside down over a freshly inoculated Biolog FF plate. An uninoculated PDA plate was used in the control treatment. The plates were kept together in place by the mean of clamps. Every 24 hours the clamps were removed, and readings of the FF plates were performed, while during the process the VOCs provider plate was kept in a cabinet flow closed by the plate lid. The PM results clearly showed stimulation effects of *C. globosum* VOCs on *M. polyspora* metabolic activity in several substrates, while except for a few substrates no effects due to *M. polyspora* VOCs were observed on *C. globosum*.

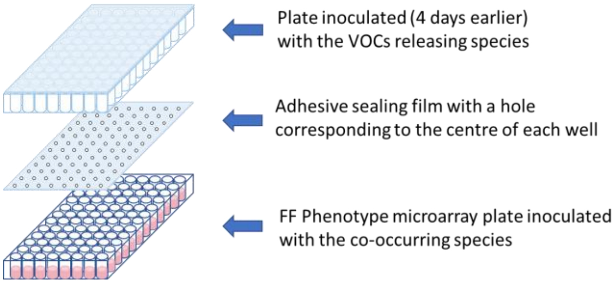

Figure 1 VOC-PM system experimental configuration

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Fungus and plastic interaction: novel results from sub-micron X-ray microscopy.

Marta Elisabetta Eleonora Temporiti[1], Flavio Cognigni[2], Lidia Nicola[1], Nicolas Gueninchault[3], Solveig Tosi[1], Marco Rossi[2]

[1]Laboratory of Mycology, Department of Earth and Environmental Sciences, University of Pavia, 27100 Pavia (Italy); [2]Department of Basic and Applied Sciences for Engineering (SBAI), University of Rome LA SAPIENZA, 00185 Rome, Italy; [3]Carl Zeiss X-ray Microscopy, Inc, 5300 Central Parkway, Dublin, CA, USA 94568;

The term plastic refers to a range of synthetic long-chain polymeric molecules which started a rapid development since 1950s, with a global production of 390 Mt in 2021. Many of the advantages for which plastic is widely used, such as its durability and hydrophobicity, then become disadvantages when a plastic product becomes waste. Indeed, they take a long time to decay and dumping them in landfills or into the environment results in the accumulation and persistence of this waste, which can also release harmful chemicals. An interesting possibility to overcome these problems is the bioremediation, which is a complex process of physic and chemical modification of polymers into smaller units mediated by microorganisms. Fungi are often involved in biodegradation processes thanks to their ability to penetrate substrates and release powerful catabolic exoenzymes. For these reasons, increasingly numerous scientific studies are focusing on the use of fungi for plastic degradation. However, the actual interaction between fungi and plastic is difficult to evaluate and assess with traditional methods. The aim of this work was to use, for the first time, high-resolution x-ray microscopy (XRM) to investigate how fungi can grow inside plastic fragments through visual substrate modifications.

A strain of *Fusarium verticillioides* was grown on polyethylene terephthalate (PET) fragments as sole carbon source for 90 days and then these fragments were analysed by XRM. Moreover, the obtained images were used as a driving tool for correlative investigations combining Raman spectroscopy, Scanning Electron Microscopy (SEM) and Energy Dispersive X-ray Spectroscopy (EDX).

The use of these techniques allowed a comprehensive understanding of the three-dimensional morphology of the fungus-plastic interaction, including elemental information and polymeric composition. In particular, XRM analysis resulted in a three-dimension al representation of the fungal growth and its distribution over the plastic substrate, highlighting also the point of fungal hyphae penetration and the creation of a fracture in PET fragments. Thanks to the ability of XRM analysis to discriminate among different densities in the samples and subsequent tests using EDX spectroscopy, it was possible to locate the mycelium, the biodegraded plastic and finally the salt crystals of cultural medium. Moreover, the fungal action on PET fragments were evaluate using Raman spectroscopy, that showed variation on the crystallinity properties of plastic and the increase in trans-glycolic units in correspondence of the fractures created by the fungal strain.

In conclusion, X-ray microscopy provided novel and surprising results regarding the geometry and morphology of the interaction between fungus and plastic substrate at sub-micrometric scale and can be used as a driving tool for further correlative analysis.

Global contribution of pelagic fungi to organic matter degradation in the ocean.

Federico Baltar[1], Zihao Zhao[1], Eva Breyer[1]

[1]Fungal & Biogeochemical Oceanography Group, Department of Functional & Evolutionary Ecology, University of Vienna, Djerassi-Platz 1, 1030, Vienna, Austria, federico.baltar@univie.ac.at

Fungi are important degraders of organic matter responsible for reintegration of nutrients into global food chains in freshwater and soil environments, but not much is known about their ecological and biogeochemical role in the open ocean water column environment. Recent evidence suggests that they are ubiquitously present in the oceanic water column (1). However, their role in processing organic matter in the ocean in comparison with that of prokaryotes is lagging behind. Here, we performed a global-ocean multi-omics analysis of all fungal-affiliated peptidases (main enzymes responsible for cleaving proteins) and CAZymes (main enzymes involved in the utilization of carbohydrates) (2,3). Together, proteins and carbohydrate, constitute the major fraction of marine living and detrital biomass. We determined the abundance, expression, diversity, taxonomic affiliation, and functional classification of the genes encoding all pelagic fungal peptidases from the epi- and mesopelagic layers. Dothideomycetes are the main fungi responsible for protease and CAZyme activity in the surface layers, whereas Leotiomycetes dominate in the mesopelagic realm. Fungal CAZyme and proteases gene abundance, diversity, and expression increased with increasing depth. This contrasts with the total occurrence of prokaryotic peptidases and CAZymes which are more uniformly distributed in the oceanic water column, suggesting potentially different ecological niches of fungi and prokaryotes. Our findings reveal a widespread active role of oceanic fungi in the cycling of C and N, highlighting potentially different ecological niches occupied by fungi and prokaryotes in the global ocean.

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Hericium erinaceus modulates gut microbiome-brain axis adapting gut microbiome composition and reducing neuroinflammation in frail aged mice.

Daniela Ratto[1], Erica Cecilia Priori[1], Fabrizio De Luca[1], Elena Savino[2], Elisa Roda[3], Paola Rossi[1]

[1]Department of Biology and Biotechnology "L. Spallanzani", University of Pavia, 27100 Pavia, Italy, daniela.ratto@unipv.it, ericacecilia.priori@unipv.it, fabrizio.deluca@unipv.it; [2]Department of Earth and Environmental Sciences, University of Pavia, 27100 Pavia, Italy, elena.savino@unipv.it; [3]Laboratory of Clinical & Experimental Toxicology, Istituti Clinici Scientifici Maugeri IRCCS, 27100 Pavia, Italy, elisa.roda@icsmaugeri.it

All living organisms naturally undergo aging, which determines the deterioration of several physiological processes (1). This progression occurs at different rates in different individuals, resulting in a heterogenous aging progression. In older adults, frailty is a geriatric syndrome that is linked to negative health outcomes, cognitive decline, and reduced quality of life. Although it is related to aging, frailty is not necessarily reflective of an individual's chronological age, highlighting the heterogeneity of the aging process among different individuals (2). Some authors, i.e., Jackson et al. and Ticinesi et al. (3,4,5), described that frailty is associated with gut microbiota dysbiosis. Indeed, more and more research are focusing on the importance of gut microbiome-brain axis in aging and frailty condition. The medicinal mushroom *Hericium erinaceus* is known for its longevity-enhancing properties (6,7) and various health benefits, especially in relation to the Central Nervous System (CNS) (8). This study investigated the effects of the oral supplementation with an Italian strain of *H. erinaceus* (He1) in frail elderly mice. Specifically, we supplemented frail mice for two months (from 21.5 to 23.5 months) with a blend of He1 mycelium and sporophore, at a content of 1 mg/die, mimicking human supplementation dose. Firstly, we measured the amount of nootropic (erinacine A, hericenones C and D) and antioxidant (L-ergothioneine) molecules in He1 extracts using HPLC-UV-ESI/MS and specific standards. Next, in frail mice we performed spontaneous behavioral tests (Emergence and Novel Object Recognition Tasks) and found that He1 supplementation significantly increased recognition memory performances during aging, partially reverting their cognitive decline. Following, we investigated, in the dentate gyrus of hippocampus and in the cerebral cortex, the cellular and molecular mechanisms at the basis of the CNS effect by analyzing specific inflammatory markers, such as interleukin-6 (IL-6) and Glial Fibrillar Acid Protein (GFAP). Two months, He1 oral supplementation significantly reduced the neuroinflammation in the investigated areas.

Next, we investigated the gut microbiome-brain axis, by addressing the prebiotic potential of *H. erinaceus.* We demonstrated that He1 oral supplementation did not change the overall gut microbiome composition, but significantly altered the relative abundance of six bacterial genera (*Odoribacter*, *Clostridia_ vadinBB60_ group, Muribaculaceae, Eubacterium_ xylanophilum_ group, Lachnospiraceae_ NK4A136_ group*, and *Clostridia_UCG-014*) specifically involved in inflammation and neuroplasticity. Altogether, these results demonstrated that *H. erinaceus* is an excellent candidate to improve recognition memory decline during aging, also through the modulation of gut microbiome composition, that could determine the reduction on neuroinflammation.

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How does phylogenomics help us to understand fruiting body evolution?

Torda Varga[12], László G. Nagy[2], Ester Gaya[1]

[1]Comparative Fungal Biology, Royal Botanic Gardens, Kew, Richmond, TW9 3AE, United Kingdom, t.varga@kew.org; [2]Synthetic and Systems Biology Unit, Institute of Biochemistry, Biological Research Center, Eötvös Loránd Research Network (ELKH), Temesvári krt. 62, Szeged, H-6726, Hungary

Fungi have been playing a pivotal role in ecosystems for millions of years, but their evolution is poorly understood. One of the reasons for the lack of knowledge is the spare fungal fossil information that hinders studying the macro-evolution of Fungi. Phylogenetic comparative methods (PCM) and phylogenomics could fill this gap by inferring the age of lineages using molecular clock methods and reconstructing the evolution of fungal traits by combining the present phenotypes and phylogenetic patterns.

An increasing number of studies have been carried out in the past years to reconstruct the major evolutionary events of various fungal lineages. Here, I summarized recent years' efforts to study the evolution of mushroom-forming fungi (Agaricomycetes), the pitfalls of phylogenomics and PCM, and how certain burdens could be overcome by whole genome sequencing of fungarium specimens.

The mushroom-forming fungi consist of species with the most diverse morphologies and various ecological traits that could affect their evolution. We reconstructed the phylogeny of 5,248 species by involving the analysis of 105 genomes and found older ages for the major lineages than before. We also inferred the speciation rate of species through time that resulted in a burst of speciation at the end of the Jurassic period that coincided with the evolution of complex fruiting bodies. Thus, we further dissected the trait-dependent speciation rate and found that traits such as the presence of the cap, velums, or having a complex hymenophore, like gills or pores, independently increased the speciation rate of lineages.

Despite the constantly improved PCM methods, many difficulties, such as incomplete taxon sampling or incorrect time inference, should be addressed when designing and carrying out such analyses. One of the critical assumptions of speciation rate models is the analysis of all existing species of the focal taxon. This assumption could be easily violated, resulting in misleading conclusions. Moreover, the molecular clock time estimates of fungal lineages have huge uncertainties ranging from tens to even hundred million years covering multiple geological periods.

Some of these difficulties could be mitigated by sampling species from underrepresented taxa and by carefully designing and carrying out molecular clock analyses. By sequencing type specimens, including Darwin's collection in the fungarium of the Royal Botanic Garden, London, we aimed to fill the gaps and improve the time estimates of the fungal tree of life, providing a backbone and reference point for future studies.

In search of the fungal plastic degraders.

Dorota Wiktorowicz[1], Agnieszka Dąbrowska[2], Julia Pawłowska[3]

[1]Institute of Evolutionary Biology, Biological and Chemical Research Centre, Faculty Of Biology, University Of Warsaw, Żwirki i Wigury 101, 02-089 Warsaw, Poland, d.wiktorowicz@student.uw.edu.pl; [2]Laboratory of Spectroscopy and Intermolecular Interactions, Faculty of Chemistry, University of Warsaw, Pasteura 1, 02-093 Warsaw, Poland, adabrowska@chem.uw.edu.pl; [3]Institute of Evolutionary Biology, Biological and Chemical Research Centre, Faculty Of Biology, University Of Warsaw, Żwirki i Wigury 101, 02-089 Warsaw, Poland, julia.z.pawlowska@uw.edu.pl

Plastics pollution has become one of the major global problems. Annual plastics global production exceeds 350 million tons, and 5–13 million tons of plastic waste are released into the marine environment (1). Various types of plastics are used, each with different properties due to different additives. The main types that dominate household waste are polyethylene, polypropylene, and polystyrene. Plastics not only can directly kill marine animals, such as turtles and seabirds but also after disintegration are a threat to the whole ecosystem, in the form of microplastics, which can make their way into food chains. Microplastics can be defined as plastic particles smaller than 5 mm, and are found in both terrestrial and aquatic environments. One of the challenges is to find methods to remove them from those environments as they are considered non-degradable, which means they need several years to degrade. Methods of plastic degradation can be divided into two main groups: biotic (biodegradation) and abiotic (e.g. thermal, chemical, photodegradation). During abiotic degradations, some toxic components can be released into the environment, it is also energy costly. In contrast to this, biotic methods should be pollution free and cheaper, with less negative impact on the environment. Currently, hundreds of organisms are reported as capable of the degradation of different plastics. Those organisms are mainly bacteria, but the list includes over 200 fungal isolates too (2). The abilities of two fungal isolates to degrade microplastics were tested. *Pyrenophora variabilis* Hern.-Restr. & Y. Marín and *Ochrocladosporium elatum* (Harz) Crous & U. Braun strains were incubated in the liquid mineral medium at room temperature with different plastic particles: polystyrene (PS) 150-250 nm, PS 700-900 nm, granulated PS, PET (glitter), granulated polyethylene (PE) and PE (rinsed out from commercial cosmetics). After 12 weeks of incubation, samples were examined. In all cases, hyphae were attached to particles, which is the first step in the biodegradation process. Although changes in pH were not observed, Raman spectroscopy analysis showed changes in the chemical structures of polymers, suggesting plastic degradation. In further experiments on those isolates, respiratory tests will be performed, and an increase in biomass will be measured. The ability to degrade complex polymers such as lignin will be tested, with the use of media containing those polymers as the only carbon source. Both *P. variabilis* and *O. elatum* belong to class *Dothideomycetes*, where a few plastic-degrading fungi are already known. Most of the *Dothideomycetes* representatives are saprobes. Among fungi that can degrade plastic, saprotrophic species and plant pathogens were reported, which is consistent with current knowledge about enzymes involved in this process (2). Overall, fungi seem to be promising candidates as they release a variety of extracellular enzymes. The two biggest current challenges are finding fungi that are the most efficient in plastics degradation and characterization of their metabolic pathways and enzyme production, which means that any research on this topic is valuable, and we should report every case of fungal plastic degradation.

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Isolation of the *Mucoraceae* from the dry-aged beef and their evaluation as potential biostarter for meat ageing.

Grzegorz Ostrowski*[1], Karina Dukaczewska[2], Danuta Jaworska[2], Magdalena Płecha[3], Wiesław Przybylski[2], Piotr Sałek[2], Krzysztof Sawicki[4], Julia Pawłowska[1]

* g.ostrowski@uw.edu.pl
1 Institute of Evolutionary Biology, Faculty of Biology, Biological and Chemical Research Centre, University of Warsaw, Żwirki i Wigury St. 101, 02-089, Warsaw, Poland
2 Department of Food Gastronomy and Food Hygiene, Institute of Human Nutrition Sciences, Warsaw University of Life Sciences (WULS), Nowoursynowska St. 166, 02-787, Warsaw, Poland
3 Institute of Biochemistry and Biophysics PAS, Pawińskiego St. 5a, 02-106, Warsaw, Poland
4 CHRIS Krzysztof Sawicki, Szwedzka St. 23/75, 30-324, Cracow, Poland

Keywords: *Mucor*, meat, maturation, fungal diversity

Dry ageing is a historically relevant method of meat preservation currently used as a way to produce high-quality beef known for its pronounced flavour and increased organoleptic qualities. Partially responsible for the taste of the dry-aged beef (DAB) may be the various microorganisms that grow on the surface of the meat. Historically, the fungal species colonising the DAB were the most often described as members of the *Thamnidium* and *Mucor* genera. In this study we used both culture based approach as well as ITS2 rDNA metabarcoding analysis to investigate the mucoralean fungal community of the DAB. The isolated fungi were members of 6 different species, belonging to the *Mucor* and *Helicostylum* genera. Metabarcoding data provided supplementary information regarding the presence of other fungi, including those from the *Thamnidium* genus. In both approaches strains closely related to the *Mucor flavus* CBS 992.68 dominated. This strain was chosen to be used as biostarter that could positively influence the quality of DAB and stabilise the dry-ageing process. The results showed a significant effect of the fungal biostarter on pH increase (0.25 units), and light myosin chain proteolysis (approximately 16%) as well as improvement of sensory quality. All together, the *M. flavus*-based biostarter was shown to increase the quality of DAB.

Latent pathogens triggered by climate change are causing novel large-scale tree decline and mortality in California.

Matteo Garbelotto[1]

[1] Department of Environmental Science, Policy and Management, University of California, Berkeley, CA 94720, USA
matteog@berkeley.edu

Large-scale tree mortality is thought to be caused by the emergence of invasive pathogens or by outbreaks of secondary organisms such as insects. Conversely, variability in resilience to climate-driven stressors may limit the scale of mortality driven exclusively by climate change. Until recently, disease caused by latent pathogens, i.e. pathogens that alternate between an endophytic, a pathogenic and a saprobic phase, has been thought to occur only in localized situations of acute stress. Since 2015, large-scale mortality of trees and shrubs, both native and exotic, has been occurring throughout Northern California. Isolations prompted by requests of various landowners, each reporting the emergence of novel die-backs, has revealed that disease of each of eight tree/shrub species studied was associated with the widespread presence of latent pathogens in the genera *Botryosphaeria*, *Diplodia*, *Neofusicoccum*, *Dothiorella*, *Diaporthe* and *Pseudosysdowia*. Although most of the fungi identified are generalists, different pathogen species were dominant and widespread on different tree species. For unreported pathogen x host combinations, inoculation studies confirmed these fungi can cause disease, although at different rates, and showed that lack of water and increasing temperatures, can lead to higher disease severity. Together, our field and lab data suggest that physiological stress caused by changing climate is triggering true outbreaks of latent pathogens, regarded now as an additional class of agents causing large scale mortality in tree populations at the regional scale.

Lichen mycobionts and endemism are key drivers for bacterial diversity associated to the thalli.

Gerardo Antonio Stoppiello[1], Roberto De Carolis[2], Claudia Coleine[1], Lucia Muggia[2], Laura Selbmann[1]

[1]University of Tuscia, Department of Ecological and Biological sciences Largo Dell'Università, 01100 Viterbo, Italy, stoppiello@unitus.it.
[2]Department of Life Sciences, University of Trieste, via L. Giorgieri 10, 34127 Trieste, Italy, decarolisrobertoce@gmail.com

In the ice-free areas along the coasts of continental Antarctica, bare rocks are the main substratum for life. Here, lichens develop very inconspicuous thalli hampering a straightforward species recognition and represent an important and prominent group of organisms, mostly endemic, also involved in primary production. Lichen thalli serve as open structures for housing a diverse range of prokaryotes, algae, and microfungi. These microcosms play a crucial role in the intricate symbiotic associations within lichens, challenging the traditional concept of lichens solely consisting of fungi (the mycobionts) and phototrophic microorganisms (the photobionts). Recently, phylogenetics, metabarcoding and 'omics' analyses, culture experiments and microscopy studies started to uncover the diversity of the lichen-associated bacterial communities. This work is part of the THALLI project, which aimes at capturing the diversity of fungi and bacteria residing within lichen thalli as hotspots for microbial diversification in one of the harshest environment on Earth. The present study focussed on bacterial diversity associated to selected lichen species to elucidate i) which taxa characterize the endemic lichen symbioses, and ii) which taxa are peculiar of extreme environmental conditions. A targeted metabarcoding was performed on five endemic species (*Acarospora flavocordia*, *Buellia frigida*, *Lecanora fuscobrunnea*, *Lecanora physciella*, *Lecidea cancriformis*) and three cosmopolitan species (*Pleopsidium chlorophanum*, *Rhizoplaca melanophthalma*, *Rusavskia elegans*). We collected a dataset of 137 samples along Northern Victoria Land over an area spanning about 450 km distance. We analyzed bacterial diversity by amplifying the variable V4 region of the 16S rRNA gene. For the first time in these lichen species and in all samples, it was thus possible to highlight a high alpha and beta diversity of bacteria taxa. These results further strengthen the perception that lichen symbioses are characterized by multiple, peculiar and ecologically related bacterial communities, acting as niches for microbial diversification and evolution in extreme environments. Moreover, we found that in the endemic lichen species bacterial diversity was significantly higher than in the cosmopolitan ones. Our findings clearly revealed that, despite the vast area sampled, bacterial diversity was primarily influenced by the species of the lichen mycobiont (i.e. the lichen species) to which they are associated, rather than the environmental conditions or the distances over which the same lichen species was collected. In conclusion, lichen species play a pivotal role in shaping bacterial communities associated to them. Such a remarkable finding emphasizes the need to consider the intricate ecological relationships when studying microbial diversity in extreme environments. The endemic lichens and their associated bacteria communities had likely benefitted of long-term coevolution under the pressure of Antarctic conditions.

Metabolomic analysis and evaluation of nutritional compounds of a new mycoprotein from artificial fungi consortia with seaweed-derived fungi and terrestrial filamentous fungi.

Catalina Landeta [1], Nicolás Salas[2], Valentina Cifuentes[1], Lhaís Araújo-Caldas[3], Allison Leyton[4], Patricia Sartorelli[3], María Elena Lienqueo[1]

[1]Center for Biotechnology and Bioengineering (CeBiB), Department of Chemical Engineering, Biotechnology, and Materials, University of Chile, Beauchef 851, Santiago, Chile.cmlandeta@uc.cl
[2]Universitat Wien, Department of Functional and Evolutionary Ecology, Vienna, Austria.
[3]Instituto de Ciências Ambientais, Químicas e Farmacêuticas, Universidade Federal de São Paulo, Diadema, SP, Brazil.
[4]Center for Biotechnology and Bioengineering (CeBiB), Center of Food Biotechnology and Bioseparations, BIOREN and Department of Chemical Engineering, Universidad de La Frontera, Francisco Salazar 01145, Temuco 4780000,

The production of animal origin proteins to satisfy human needs require abundant natural resources and also their intake is associated with health problems (1). Mycoprotein is a unique a nutritious protein source derived from the filamentous fungi. In this research, we obtained for the first time an alternative and functional protein food from an artificial fungi consortium with marine and terrestrial filamentous fungi. Additionally, this product is the result of enhancing the nutritional value from the Chilean brown seaweed *Durvillaea* spp. through a submerged fermentation process with our consortium designed. This is regarded as part of the solution to debottleneck the physiological limitations of pure cultures bioprocesses, such as degrading complex carbon sources and efficient substrate utilization (2). This rationally designed consortium demonstrated properties that exceeded the pure culture properties, including a ~150% increase in productivity, an increase in total protein (~336%), amino acids (~245%), ß-D-glucans (100%), and total (Poy)phenols (75%). This mycoprotein has all the essential amino acids, low content of fatty acids (3%), high level of dietary fibre (35%), high antioxidant activity (TEAC of 34 µM/g), good concentration of (Poly)phenols (1,4 mg/g EAG), without mycotoxin, heavy metals and pesticides. The nutritional composition of mycoprotein was evaluated, obtaining a yield of 561.3 g of mycoprotein from 1 kg of dry seaweed, a good concentration of total protein (35 g/100 g dw), a considerable concentration of total amino acids (21 g/100 g dw), and good concentration of ß-D-glucans (22g /100g dw). In this study, an untargeted metabolomics approach combined with multivariate statistical analysis and dereplication techniques aides by the GNPS Molecular Network was employed to screen the metabolites and to identify molecules with nutraceutical properties. Among the screened 110-top differential metabolites, 22 nutraceutical compounds showed higher content in co-cultive (trehalose, oleamide, linoleic acid, omega-3 (ω-3) polyunsaturated fatty acid (PUFA), ergosterol, among others). Additionally, the functional properties of this mycoprotein were evaluated. The properties to be considered were: antioxidant (radical eliminator DPPH and ABTS), anti-obesity, anti-diabetic. Inhibition of pancreatic lipase, α-glucosidase, and α-amylase enzymes was used to determine anti-obesity and anti-hyperglycemic activity. In conclusion, the mycoprotein from artificial fungi consortia through a process of submerged fermentation with *Durvillaea* spp. as sole carbon sources, results in a product that has a high-quality protein, great nutritional value, with prebiotic and nutraceutical potential. Also we found an increase in the diversity of mycochemical compounds which represents a new source of drugs, nutraceuticals and functional food.

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Metabolomics as a tool to discriminate *Ganoderma lucidum* and *Ganoderma valesiacum* mycelium grown in liquid culture medium.

Gaia Cusumano[1], Giancarlo Angeles Flores[2], Paola Angelini[1], Luca Candelaresi[1], Roberto Maria Pellegrino[1], Carla Emiliani[1], Roberto Venanzoni[1]

[1] Department of Chemistry, Biology and Biotechnology, University of Perugia, 06122 Perugia, Italy; gaia.cusumano@studenti.unipg.it; paola.angelini@unipg.it; luca.candelaresi@studenti.unipg.it; roberto.pellegrino@unipg.it; carla.emiliani@unipg.it; roberto.venanzoni@unipg.it
[2] Botanic Garden "Giardino dei Semplici", Department of Pharmacy, "Gabriele d'Annunzio" University, Via dei Vestini 31, 66100 Chieti, Italy; giancarlo.angelesflores@unich.it

Ganoderma P. Karst. (phylum *Basidiomycota*) is a highly diverse and globally distributed genus known for causing white rot wood decay in various tree species. For the past century, studies on *Ganoderma* species in North America and other regions have often referred to any laccate *Ganoderma* species as *G. lucidum* (Curtis) P. Karst. *sensu lato*. However, molecular research has now confirmed that *G. lucidum sensu stricto* is native to Europe and certain parts of China. *G. lucidum* basidiocarps are predominantly found on oak trees but can also be observed on other tree species. In Europe, they have been documented on 24 different woody plant genera (1). *Ganoderma valesiacum* Boud. can sometimes be mistaken for *G. lucidum* or *G. carnosum* Pat., but it can be differentiated by its very short or rudimentary stipe and its often-cracked laccate crust, which exposes a white context (2). However, the taxonomic status of *G. valesiacum* remains uncertain due to unclear relationships with *G. lucidum* (2). The main objective of this study is to investigate the relationship between taxonomic markers (ITS and RPB2) and the metabolomic profiles, through LC/MS-QTOF method, of *G. lucidum* and *G. valesiacum* mycelium cultivated in a liquid culture medium. The results revealed that the cladogram derived from the metabolic profile of the mycelia clearly distinguished *G. lucidum* from *G. valesiacum*. Among the metabolites that distinguished *G. valesiacum* from *G. lucidum* we found: 8,11,14-Eicosatrienoyl-coA and Glucosamine 6-phosphate, involved in several metabolic pathways including fatty acid and amino acids metabolism. Conversely, the main metabolites distinguishing *G. lucidum* from *G. valesiacum* were: Homocitric and Isocitric Acid, involved in the metabolism of glyoxylate and dicarboxylate and in the citrate cycle (TCA cycle).

This study suggests that metabolic profile analysis, together with morphological and molecular analysis, could be useful in distinguishing *G. valesiacum* from *G. lucidum*.

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Fungal conservation: Inoculation of heart-rot in living trees – establishing heart-rot fungi for translocation and veteranisation.

Matthew Wainhouse[1] & Lynne Boddy[1]

[1] Cardiff University, Cardiff, UK/ Natural England, Bristol, UK, matthew.wainhouse@naturalengland.org.uk
[2] Cardiff University, Cardiff, UK, Boddyl@cardiff.ac.uk

Heart-rot and cavities in living trees are major drivers of tree associated biodiversity. A long history of forestry and land-use change has created a fragmented treescape with a depleted stock of older, 'veteran' trees that support these microhabitats. Heartwood decomposition is a slow process, and it may take heart-rot fungi hundreds of years to establish in long lived trees before hollowing even begins. This has left an age gap in the development of heart-rot and hollowing which is damaging to rare heart-rot fungi and cavity dependant taxa. A major challenge in forest and wood pasture restoration is how heart-rot processes can be accelerated or replicated in younger trees. One approach that we have been trialling is to directly inoculate trees with heart-rot fungi. Here we present the early results from two long-term heart-rot inoculation trials: (1) A Veteranising inoculation designed to benefit cavity and decay dependant fauna using 4 species; and (2) Translocation inoculation, to reintroduce 2 locally extinct, dispersal-limited heart-rot fungi. In both trials we were able to demonstrate the successful establishment of heart-rot fungi. Nonetheless there have been challenges and failures. We share results from these trails and insight on designing and monitoring conservation inoculations. Conservation inoculations have the potential to be a useful tool in forest restoration and we hope to stimulate wider uptake as a direct method for conservation.

Microwave-assisted extraction and medicinal mushrooms: an innovative and promising technique with significant applicative implications.

Valeria Ferraro[1], Maria Letizia Gargano[2], Natalie Paola Rotondo[1], Annalisa De Palma[3], Giovanni Lentini[1], Maria Maddalena Cavalluzzi[1]

[1]Department of Pharmacy-Pharmaceutical Sciences, University of Bari “Aldo Moro”, University Campus “Ernesto Quagliariello”, via E. Orabona, 4, 70125 Bari, Italy; valeria.ferraro@uniba.it; natalie.rotondo@uniba.it; giovanni.lentini@uniba.it; mariamaddalena.cavalluzzi@uniba.it; [2]Department of Soil, Plant, and Food Sciences, University of Bari “Aldo Moro”, Via Amendola 165/A, I-70126 Bari, Italy; marialetizia.gargano@uniba.it; [3]Department of Biosciences, Biotechnologies, and Environment, University of Bari “Aldo Moro”, via E. Orabona, 4, 70125 Bari, Italy; annalisa.depalma@uniba.it

There are an increasing number of nutraceutical and mycotherapeutic products in which mushrooms are included as active ingredients due to their content of multiple bioactive compounds responsible for innumerable medicinal and health-promoting properties. Medicinal mushrooms can be used as either whole carpophore powder or extract, or as a combination of both. Nowadays, both research and production processes must adhere to the principles of Green Chemistry, whose keystones include optimizing energy and resource consumption, as well as minimizing the use and/or generation of hazardous substances and wastes. Therefore, especially with a view to a standardized, high-quality industrial production, it is essential to adopt efficient extraction techniques both in terms of yield and quality of the obtained product, and in terms of cost and environmental sustainability. One of the currently most innovative and promising techniques for obtaining metabolites from plant matrices is the microwave-assisted extraction (MAE) (1), but to date only a few studies report its application to mushrooms (2, 3, 4, 5). We therefore carried out an investigation comparing MAE with other techniques, innovative or traditional, in terms of both efficiency and effect on antibacterial and antidiabetic activity of medicinal mushrooms extracts. This study provides evidences that MAE is consistently advantageous compared to other methods in that it allows a higher extraction yield and/or a significant reduction in the required time and solvent volumes, as well as in some cases an enhancement of the bioactivity of the extract. These results are promising both for more in-depth research into the activities of medicinal mushrooms and their bioactive metabolites, and for a potential future industrial application of MAE to obtain high-quality nutraceutical and mycotherapeutic formulations with a view to improved sustainability and cost reduction.

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Mucoromycota-bacterial interactions in postindustrial soil.

Beniamin Abramczyk[1], Julia Pawłowska[1]

[1]Institute of Evolutionary Biology, Faculty of Biology, Biological and Chemical Research Centre, University of Warsaw, ul. Żwirki i Wigury 101, 02-089 Warsaw, Poland, b.abramczyk2@student.uw.edu.pl

Fungi of *Mucoromycota* phylum are known to frequently interact with bacteria [1,2]. Bacteria can alter the metabolism of their host affecting its capacity of different environment colonization [3,4]. Information about these interactions in different environments could be used in neutralizing soil pollution (e.g. with hydrocarbons), which is a serious threat to soil functioning all over the world [5]. We aimed to examine the frequency of *Mucoromycota*-bacterial interactions in postindustrial soil. Soil was sampled in places where elevated concentrations of hydrocarbons were expected. Samples were also taken from control sites located nearby. Concentrations of the above-mentioned pollutants were measured in the samples using gas chromatography with a mass spectrometer (GC–MS) for qualitative analysis and gas chromatography with a flame ionization detector (GC-FID) for determining the total amounts of petrol, mineral oils, and PAHs. *Mucoromycota* strains were isolated using Warcup method [6]. They were identified molecularly based on ITS rDNA sequence analysis [7]. Then bacterial presence was examined by PCR amplification of 16S bacterial rDNA [2]. Taxonomic composition of five chosen species microbiomes was examined using V3-V4 16S bacterial rDNA fragment amplification and sequencing on Illumina platform. Amplicon data was filtered and classified using QIIME2 [8] and Silva database. Bacteria were detected in 32% of isolated *Mucoromycota* strains. Some of the strains in which bacteria were found were classified as *Mucor hiemalis* (previously not reported to interact with bacteria). Frequency of detected bacterial-fungal interactions was positively correlated with elevated levels of aliphatic low molecular weight hydrocarbons (C6-C12) and mineral oil in soil (p-value derived from chi^2 test < 0.01). It is probably the consequence of high selection pressure in soil contaminated with these pollutants, which promotes fungal-bacterial interactions. Regardless of the place of isolation and species, *Burkholderiaceae* family members were detected in their microbiome. In four out of five examined species (including *Mucor hiemalis*), *Burkholderia-Cabaleronia-Paraburkholderia* clade representatives were detected. Representatives of this clade are well known to form endohyphal interactions with Mucoromycota fungi [2]. Consequently this data is an indirect proof of *Mucor hiemalis* and other Mucoromycota isolates harboring endohyphal bacteria. Other abundant bacterial genera were *Serratia*, *Pseudomonas* and *Paenibacillus* also reported previously to interact with fungi [9]. *Pseudomonas* dominated in the microbiome of species found in soil with extremely high hydrocarbon concentrations. *Pseudomonas* representative is known to enhance hydrocarbon degradation capacity of *Mucoromycota* representative [10]. Consequently bacteria of this genus could be recruited by fungi and increase their fitness in such polluted environments. Nevertheless further research is required to prove this hypothesis.

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Mushrooms cultivation: current status and future perspectives

Federico Puliga[1], Veronica Zuffi[1], Yue Huang[1], Ornella Francioso[1], Alessandra Zambonelli[1].

[1]Department of Agricultural and Food Sciences, University of Bologna, Viale G. Fanin 40-50, 40127, Bologna, Italy.

Mushrooms represent one of the oldest foods for humans and have been harvested and cultivated for millennia. They are considered an important source of nutrients and biologically active compounds with several medicinal properties.

Mushroom cultivation is widespread all over the world, with a global volume production of around 40 million tons, and China is the largest producer (1). More than 60 species of saprotrophic fungi are commercially cultivated (2), and the principal genera are *Lentinula*, *Pleurotus*, *Auricularia*, and *Agaricus* (3). Edible mushrooms are rich in proteins (15-35%), carbohydrates (35-70%), fibers, vitamins, minerals, and low in fat (< 5%) (4,5). Moreover, several species such as *Ganoderma lucidum* (Curtis) P. Karst., *Lentinula edodes* (Berk.) Pegler, *Grifola frondosa* (Dicks.) Gray, *Hericium erinaceus* (Bull.) Pers., *Agaricus blazei* Murrill, and some *Pleurotus* species, have received attention due to their antimicrobial, anti-inflammatory, immunomodulatory, antidiabetic, cytotoxic, antioxidant, hepatoprotective, anticancer, antioxidant, antiallergic, antihyperlipidemic, and prebiotic properties (6).

Cultivation of edible mushrooms is becoming increasingly successful in many countries since it is not so impactful considering the energy input, the land usage, and the growth time required. Several edible and medicinal species are in the large-scale production, and thanks to use of smart farming systems all the environmental parameters can be monitored, and the production automatically harvested. Lignocellulolytic mushrooms such as oyster mushrooms, and shiitake are typically cultivated on different pasteurized or sterilized lignocellulosic substrates. Most of residual biomasses from agriculture are rich in lignin, cellulose and hemicellulose and thus, they can be used for the cultivation of these mushrooms and recycled sustainably according to circular economy. As a result, mushroom companies are beginning to re-evaluate their production processes. Most notably, residual biomasses from agriculture differing from those commonly used in the production process have been explored in the recent decades, with the aim of reducing costs and, at the same time, enhancing the value of by-products from other supply chains (7). In this context, the status and the future perspectives of the mushroom cultivation industry are explored.

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Mycological Groups & Societies in Europe: how can we cooperate?

Izabela Kałucka[1], Alessandra Zambonelli[2], Solveig Tosi[3], Julia Pawłowska[4]

[1]European Mycological Association, izabela.kalucka@biol.uni.lodz.pl; [2]Italian Union of Mycology, alessandr.zambonelli@unibo.it; [3]Italian Botanical Society working group for mycology, solveig.tosi@unipv.it; [4]Polish Mycological Society, julia.z.pawlowska@uw.edu.pl;

Although *Fungi* were recognized as a separate eukaryotic kingdom only in the middle of 20th century (1), the history of mycological groups and societies not only in Europe is much longer. Various informal groups of mushroom pickers have functioned for hundreds of years of human history. However, the 19th century, especially in Europe and Northern America, is a period of intensive development of naturalist clubs and scientific societies at regional and national levels. For example, La Société mycologique de France was created in 1884 (http://www.mycofrance.fr/), the British Mycological Society was formally established in 1896 (2), the Czechoslovak Mycological Society (www.myko.cz) and The German Society for Mycology (https://www.dgfm-ev.de/) in 1921. Currently, more than 120 mycological societies are listed on the European Mycological Association webpage (http://www.euromould.org/resources/links/socs.html). After over 100 years of independent history and development under different geopolitical and societal conditions, the landscape of mycological groups and organizations in Europe varies greatly. In some countries, several smaller mycological associations and groups function simultaneously, while in others, national-level societies dominate. Some of them are already collaborating on an international level, for example organizing regional meetings like The Nordic Mycological Congress, while others focus on national-level activities. Over the last decades, numerous efforts have been made to integrate mycological societies at the international level. Although the Congress of European Mycologists, which first took place in 1956, is the world’s oldest continuously running series of international meetings about fungi, the European Mycological Association (EMA) was founded only in 2003 during the XIV Congress of European Mycologists organized in Crimea (Ukraine) by decision of more than 100 delegates from 30 countries. It is worth mentioning that the European Council for the Conservation of Fungi (ECCF), which was founded in 1985 and gathered some 80 representatives from most European countries, has become an autonomous conservation wing of the EMA. The main objectives of the EMA are to promote the study of fungi, disseminate knowledge and information about fungi and their conservation, improve the infrastructure of mycology within Europe, and to establish and foster relations between those working towards such objectives. The EMA’s 20th anniversary is a good moment to discuss international collaboration opportunities between European societies to develop mycological research beyond national borders.

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Mycoproteins of medicinal mushroom from submerged fermentation of agroindustrial byproducts.

Davide Ferrero[1], Elisa Moscato[1], Federica Spina[1], Maria Maddalena Cavalluzzi[2], Giuseppe Mannino[3], Sara Bellezza Oddon[4], Maria Letizia Gargano[5], Giuseppe Venturella[6], Cinzia Margherita Bertea[3], Laura Gasco[4], Giovanna Cristina Varese[1]

[1]Department of Life Sciences and Systems Biology (DBIOS) *Mycotheca Universitatis Taurinensis* - Università degli Studi di Torino, Viale Mattioli 25, Torino, Italy;
[2] Department of Pharmacy - Drug Sciences, Università degli Studi di Bari Aldo Moro, Piazza Umberto I, Bari, Italy;
[3] Department of Life Sciences and Systems Biology (DBIOS) Università degli Studi di Torino, Via Quarello 15/A, Torino, Italy;
[4] Department of Agricultural, Forest and Food Sciences (DISAFA) Università degli Studi di Torino, Via Leonardo da Vinci 44, Grugliasco, Italy;
[5] Department of Soil, Plant and Food Science (DISSPA) Università degli Studi di Bari Aldo Moro, Piazza Umberto I, Bari, Italy;
[6] Department of Agricultural, Food and Forest Sciences (SAAF) Università degli Studi di Palermo, Viale delle Scienze 4, Palermo, Italy.

The global population is constantly growing and likely to exceed 10 billion by 2060[1], thus revising food production and its resources is necessary. Our planet is overexploited and 80% of Earth's agricultural land is used for livestock[2], for meat and meat derivates consumption, which have heavy environmental impact (high water consumption, production of greenhouse gases, use of territorial resources, deforestation and biodiversity loss). To overcome these problems without depriving human diet of proteins, several solutions have arisen over time, such as protein-rich plants, synthetic meat and insects. Mycoproteins can be a valid alternative. Fungi are highly prone to biotechnological applications, such as making plant matrices more digestible by degrading lignin for feed purpose, enriching plant wastes in fatty acids and proteins, including essential amino acids, and for using their biomasses as probiotics. Moreover, these processes are often coupled with production of valuable metabolites such as vitamins and pigments[3]. Although retrieving proteins from fungi has already been achieved using solid state fermentation, it arouses technological and environmental concerns. Submerged fermentation is instead a more sustainable and controlled solution, avoiding seasonal process dependance. Developing products based on this technology could be the foundation for a new 'green' food frontier. Five edible medicinal mushrooms (*Ganoderma resinaceum*, *Pleurotus ostreatus*, *Cordyceps militaris*, *Pleurotus eryngii* and *Lentinula edodes*) preserved in the *Mycotheca Universitatis Taurinensis* (University of Torino) were selected. To develop economic and energetic sustainable process, growth media were based on agroindustrial by-products (e.g. wheat vinasses, insects exuviae, tomato peels, etc.) coming from local companies, to identify the media yielding the more fungal biomass. Then, once selected the nine best performing media, the proximate compositions of the fungal biomasses were analyzed to identify the highest protein content biomass for future studies focused on fungal protein production. Among the experimental lines, both the highest biomass production (24 g/L dw) and protein content (51% dw) were achieved in *P. eryngii* grown in *Hermetia illucens* exuviae-based media. To the authors' knowledge, this is the first work exploiting insect exuviae-based media to sustain fungal growth in SmF. At the same time, antioxidant activity of the extracts retrieved from these fungal biomasses was measured. Extracts were obtained via conventional 24-h maceration or fast microwave (MW) treatment, using water or ethanol as solvents. Data indicated that MW method led to comparable antioxidant activity yields, opening new possibilities in future (poly)phenolic extraction protocols with more feasible and sustainable techniques. Studies are ongoing using different fungi and by-products in respect of the circular Economy and sustainability principles. The present project leads to a better understanding of sustainable fungal fermentation, sustainable downstream processes of valuable biomasses for their potential exploitation in food and feed sector.

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Mycoremediation in agricultural soils heavily contaminated by hexachlorocyclohexane (HCH) isomers: insights from the HCH-degrading *Penicillium griseofulvum* MUT5854.

Domenico Davolos[1], Andrea Ceci[2], Alessandra Ricelli[3], Oriana Maggi[2], Anna Maria Persiani[2]

[1]Department of Technological Innovations and Safety of Plants, Products and Anthropic Settlements, INAIL, Research Area, Via R. Ferruzzi, 38/40, 00143, Rome, Italy, d.davolos@inail.it; [2]Department of Environmental Biology, Sapienza University of Rome, Piazzale A. Moro 5, 00185, Rome, Italy, andrea.ceci@uniroma1.it, oriana.maggi@gmail.com, annamaria.persiani@uniroma1.it; [3]Institute of Molecular Biology and Pathology, CNR, Piazzale A. Moro 5, 00185, Rome, Italy, alessandra.ricelli@cnr.it

The contamination of soil by toxic organochlorine pesticides such as hexachlorocyclohexane (HCH) isomers threatens human health. The biotransformation of HCH by microfungi may be of outstanding importance for mycoremediation of HCH-polluted sites. *Penicillium griseofulvum* MUT5854, isolated from agricultural soils with high concentrations of HCH isomers, biotransformed alfa-, beta-, gamma-, and delta-HCH isomers (1) but did not show azole resistance. However, very little information exists on the genes potentially relevant to mycoremediation, such as those involved in the stress response and in the secondary metabolite (SM) biosynthesis. Modern molecular approaches can help to study the potential of filamentous fungi such as *P. griseofulvum* MUT5854 that survive in environments with a high concentration of toxic pollutants. Here, in culture filtrate of MUT5854 patulin and griseofulvin were extracted, detected and quantified by HPLC. Notably, *P. griseofulvum* MUT5854 produced more patulin than griseofulvin under lab conditions. The production of patulin, a mycotoxin that can cause health concerns, in *Penicillium* spp., is regulated by transcription factors but can be also affected by environmental factors (e.g., pH, nutrients, temperature). However, we do not know the regulatory details of patulin production in MUT5854 and whether contamination by toxic pollutants such as HCH could be involved in the regulatory network. In this study, the genome of *P. griseofulvum* MUT5854 was sequenced, assembled, and annotated (29 Mb, GC % 47.28, 12,306 genes). In the *P. griseofulvum* MUT5854 genome (BioSample SAMN29206920; BioProject PRJNA850932; SRA SRS14357487), there were candidate genes that might contribute to the HCH tolerance/resistance and are potentially involved in the biodegradation pathway for HCH (2). Moreover, analysis of the *P. griseofulvum* MUT5854 genome predicted 46 biosynthetic gene clusters (BGCs) potentially involved in the SM biosynthesis, of which many belong to non-ribosomal peptide synthase (nrps) or nrps-like, type I polyketide synthase (t1pks), and terpene, while there were few indole clusters. Among the putative BGCs, SM clusters shared homology with previously proposed BGCs and were potentially responsible for the synthesis of patulin, griseofulvin, penicillin, yanuthone D and cyclopiazonic acid. The genomic DNA sequence is the key to understanding patulin synthesis in *P. griseofulvum* MUT5854, providing insight into understanding the regulatory mechanism of patulin production, including the regulators outside the biosynthetic gene cluster. Moreover, our genomic data on *P. griseofulvum* MUT5854 provide an essential basis for transcriptomic approaches under exposure to HCH, which is critical for a better understanding of molecular mechanisms to enhance the mycoremediation experimental approaches for HCH, including *in situ* bioremediation of HCH isomers. Future work will be aimed to ascertain HCH biodegradation capability in relation to mycotoxin production of *P. griseofulvum* MUT5854 in HCH-contaminated soil microcosms.

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NEW RED DATA LIST OF LATVIAN FUNGI (AGARICALES S.L.) BASED ON IUCN CRITERA

T: Fungal conservation

Inita Dāniele

Institute of Biology, University of Latvia, Ojāra Vācieša str. 4, Riga, Latvia, inita.daniele@lu.lv,
Latvian National museum of Natural history, Kr. Barona str. 4., Riga, Latvia, inita.daniele@lndm.gov.lv

The previous Red Data List of fungi was developed as a part of the Red Data Book in 1996 (1). It contained 38 fungi species. Their conservation state assessments were based solely on expert opinion. In Latvia national legislation, requirements of protection of species and habitats are defined by the Law of Protection of Species and Habitats (ratified by the Parliament on 16.03.2000.). It contained 62 fungi species. In 2021-2023, during the development of the new Red Data List, species conservation state assessments were carried out accordingly to the standardized and worldwide used IUCN criteria (2). The outcome of this project will be an important and lasting step for fungal conservation. Currently, 112 species has been nominated to be red-listed in Latvia. 46 or 40% of them are Agaricales s. l. species. 10 species (*Amanita ceciliae*, *Butyriboletus appendiculatus*, *Chrysomphalina chrysophylla*, *Desarmillaria ectypa*, *Hygrocybe intermedia*, *Hygrocybe punicea*, *Hygrocybe quieta*, *Hygrocybe spadicea*, *Porpolomopsis calyptriformis*, *Strobilomyces strobilaceus*) are critically endangered in Latvia, 20 species (*Agaricus devoniensis*, *Amanita eliae*, *Amanita strobiliformis*, *Amanita gemmata*, *Caloboletus calopus*, *Cortinarius praestans*, *Cyanoboletus pulverulentus*, *Entoloma sinuatum*, *Hygrophorus chrysodon*, *Hygrophorus nemoreus*, *Hymenopellis radicata*, *Lactarius serifluus*, *Leccinellum crocipodium*, *Lentinus tigrinus*, *Pluteus aurantiorugosus*, *Pseudoboletus parasiticus*, *Rhodotus palmatus*, *Tricholoma acerbum*, *Tricholoma apium*, *Volvariella caesiotincta*) - in endangered species category, 16 species (*Agrocybe firma*, *Entoloma euchroum*, *Gliophorus psittacinus*, *Gyroporus castaneus*, *Hebeloma radicosum*, *Hemistropharia albocrenulata*, *Lactarius lignyotus*, *Lepiota erminea*, *Lepiota grangei*, *Leucopaxillus tricolor*, *Marasmius wynneae*, *Pholiota heteroclita*, *Pleurocybella porrigens*, *Pluteus umbrosus*, *Porphyrellus porphyrosporus*, *Volvariella bombycina*) are vulnerable. Fungi conservation state assessments were carried out accordingly to the IUCN guidelines for the Red List categories and criteria (IUCN Standards and Petition Committee, 2019). The distribution data was based on the species records, available at the Nature Conservation Agency database OZOLS, public nature observation portal dabasdati.lv, the website for the fungi of Latvia fungi.lv, and data of Latvian National Natural history museum fungi collection (nmkk.lv).

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No fungus is an island: how endohyphal bacteria may affect enzymatic capacities of their hosts?

Julia Pawłowska[1], Beniamin Abramczyk[1], Zuzanna Błocka[1], Maria Furman[1], Robert Stasiuk[2], Renata Matlakowska[2], Alicja Okrasińska[1]

[1]Institute of Evolutionary Biology, Faculty of Biology, University of Warsaw, Żwirki i Wigury 101, 02-089 Warsaw, Poland, julia.z.pawlowska@uw.edu.pl; [2]Institute of Microbiology, Faculty of Biology, University of Warsaw, Miecznikowa 1, 02-096 Warsaw, Poland.

Fungi and bacteria play an important role in soil-forming processes, as together they are responsible for the majority of decomposition processes. Nowadays, the most common approach to asses soil microbiome is metabarcoding but it does not elucidate the function of detected organisms, which is needed for a more comprehensive understanding of ecosystem functioning. *Mucoromycota* fungi, one of the most ancient groups of land fungi, commonly occur in various types of soils, including post-industrial soils contaminated with polycyclic aromatic hydrocarbons (PAHs) (Okrasińska *et al.* 2022). At the same time, these fungi are known to harbor endohyphal bacteria (EHB) making them perfect candidates for a studying bacterial-fungal interactions in soil. By combining traditional microbiological isolation techniques with metabarcoding, genomic analyses, and metabolomic physiological experiments we aimed to understand how endohyphal bacteria may affect the enzymatic capacities of their fungal hosts in anthropogenically transformed environments. The *Mucoromycota* strains from polluted and clean control sites were isolated and the presence of EHB was checked for each strain in order to assess prevalence of endohyphal bacteria in both cases. It was confirmed that the EHB are more frequent in fungal strains isolated from soils polluted with hydrocarbons. Additionally, a common and facultative interaction between bacteria from the genus *Paraburkholderia* and *Umbelopsis* sp. fungal host was discovered (Okrasińska *et al.* 2021). We observed that endohyphal *Paraburkholderia* sp. has a negative impact on the growth rate of *Umbelopsis* sp. in poor medium conditions. In order to characterize molecular mechanism of the interaction, we decided to analyze the genomes of both partners. Analysis of annotated genomes further confirms our hypothesis of the facultative nature of this symbiosis. The presence of several bacterial genes extending fungal metabolic capacities was detected. Furthermore, GC-MS (gas chromatography with mass spectrometry) analysis of wildtype and cured strains revealed differences in metabolite composition, indicating the significant role these bacteria may play in adapting their hosts to harsh environmental conditions.

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Oceanic fungal diversity and biomass distribution in the Atlantic Ocean.

Eva Breyer[1], Franz Berthiller[2], Katherine Salazar[1], Federico Baltar[1]

[1]Fungal and Biogeochemical Oceanography, Department of Functional and Evolutionary Ecology, University of Vienna, Djerassiplatz 1 A-1030 Vienna, Austria, eva.breyer@univie.ac.at; federico.baltar@univie.ac.at; [2]Institute of Bioanalytics and Agro-Metabolomics, Department of Agrobiotechnology (IFA-Tulln), University of Natural Resources and Life Sciences, Vienna, Konrad Lorenz Str. 20, 3430 Tulln, Austria

Fungi have played a significant role in shaping the biosphere since the early stages of life on Earth. While fungi can be found in various environments, most environmental research has focused on soils and freshwater ecosystems. Despite their importance as parasites during phytoplankton blooms and decomposers and recyclers of dead organic matter, our understanding of fungal communities in marine environments, particularly within the pelagic water column, remains limited. Recent analysis of a global 18S rRNA dataset shed light on fungal diversity and community composition across major ecosystems, revealing distinct patterns along temperature, salinity, and latitude gradients (1). Chytridiomycota were found to dominate coastal waters, while Asco- and Basidiomycota prevailed in oligotrophic oceans (1). Additionally, the crucial role of oceanic mycoplankton in marine carbon and nitrogen cycling has only recently been recognized (2,3). However, compared to other microbial groups considered to be important players in marine biogeochemical cycles, detailed studies investigating fungal diversity, community composition in relation to basic parameters such as the distribution of fungal biomass in major ocean basins are lacking. To address these gaps, we conducted a comprehensive study collecting oceanic fungal biomass from surface waters to the abyssopelagic zone. Our sampling covered both productive and oligotrophic environments, encompassing both hemispheres in the Atlantic Ocean, and spanning two oligotrophic gyres. Additionally, we analyzed fungal diversity and community composition. By employing ergosterol extraction and fluorescence microscopy as proxies to pelagic fungal cultures, we calculated novel biomass conversion factors applicable to future oceanic samples. Our findings revealed the highest fungal biomass in productive surface regions, which gradually decreased with increasing distance from the coast. Distinct fungal communities were observed in productive and oligotrophic waters. While Asco- and Basidiomycota dominated throughout the whole transect, Chytridiomycota exhibited higher relative abundances in productive coastal areas, potentially related to their parasitic role during phytoplankton blooms. In summary, our results highlight a substantial contribution of pelagic fungi to marine microbial diversity and biomass, underscoring the importance of integrating oceanic fungi into marine biogeochemical cycles and diversity assessments. By expanding our understanding of fungal ecology in the marine realm, we gain valuable insights into the complex interactions shaping marine ecosystems and nutrient cycling dynamics, while expanding the knowledge of the global relevance of fungi in Earth's ecosystems.

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Open access data as source for discovery.

Wieland Meyer[1]

[1]Westerdijk Fungal Biodiversity Institute of the KNAW, Utrecht, Netherlands
w.meyer@wi.knaw.nl

With the advances in molecular biology, genomics, proteomics, and transcriptomics, ever increasing amounts of data are generated. As most studies are publicly funded there is a call for open access of the obtained datasets. Journals are requiring the submission of the underlying data to public databases, such as GenBank, RefSeq, SWISS-PROT, etc. Legal obligations resulting from the Nagoya Protocol on Access and Benefit Sharing are on one site beneficial but on the other site also potentially hampering global research if also applied to data. It has been shown that all sites are equally profiting from open access data sharing. Without public access to sequence information e.g., scientific progress would be slowed down and more seriously methods as DNA barcoding or MALTI-TOF for species identification/diagnostic would be impossible. The Barcode of Life (BOLD), UNITE, RefSeq, the ISHAM barcode database and the French Spectral Library are the fundamental backbone for clinical diagnosis of fungal infections. The Sequence Read Archive (SRA) of the NCBI at the NIH, USA, is the largest sequence depository in the world. Recent studies looking at the origin of the multidrug resistant yeast *Candida auris*, coursing outbreak in several hospital settings globally or the distribution of the *Candida haemulonii* complex, highlight the hidden potential of “big data” to unlock new directions in all fields of biological research, e.g., in relation to the identification of starting points for the search of the origin and spread of emerging fungal pathogens, emphasizing the importance of archiving and curating big data and making them freely accessible to the whole scientific community.

Polar and non-polar cold habitats: an ecological niche harboring extremophilic fungal diversity.

Pietro Buzzini**[1]**, Ciro Sannino[1], Luigimaria Borruso[2], Gianmarco Mugnai[1], Alessandro Bernetti[1], Daniele Andreani[1], Benedetta Turchetti[1]

[1]University of Perugia, Borgo XX Giugno 74, Perugia, Italy, pietro.buzzini@unipg.it
[2]Free University of Bozen, Piazza Università 1, Bolzano, Italy

Polar and non-polar cold habitats harbor the life of both psychrophilic and psychrotolerant fungi, which have developed specific adaptation mechanisms to low temperatures (1, 2). Because the impact of ongoing global warming on cold habitats is intensifying every year, fungal diversity colonizing worldwide cold areas may be considered sentinel biological systems for studying the effect of climate changes on microbial diversity (2).

Soil, permafrost, brine, ice and debris samples were collected from Antarctic and Alpine sites. Fungal diversity (including filamentous and yeast life forms) was investigated via NGS targeting the ITS2 region. Bioinformatic analysis of raw sequences was performed by Qiime 2 (https://qiime2.org/). The taxonomic assemblage of fungal communities and their interaction with physical/chemical (abiotic) parameters were studied by diversity indices, linear discriminant analysis effect size (LEfSe) and Pearson correlations.

Operational Taxonomic Units (OTUs) assigned to the phylum Ascomycota predominated among filamentous fungi, while Basidiomycota dominated among yeasts. At the phylotype level, yeast life forms dominated the fungal communities: *Candida*, *Glaciozyma*, *Leucosporidium*, *Malassezia*, *Naganishia* and *Sporobolomyces* were the most frequently found genera. The analysis of diversity revealed a high phylogenetic differentiation among samples, even at the small-scale level. A few abiotic parameters (i.e. salinity, conductivity, pH and the concentration of organic C) exhibited a significant ($p < 0.05$) impact as drivers of fungal diversity. Significant ($p < 0.05$) interactions among different fungal taxa were also found.

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Potential importance of the environmental DNA for improving fungal Red Listing.

Ovidiu Copot[1], Asko Lohmus[1], Kessy Abarenkov[2], Leho Tedersoo[1], Kadri Runnel[1]

[1]Institute of Ecology and Life Sciences, University of Tartu, Tartu, J. Liivi 2, Estonia, om@ut.ee; [2]Natural History Museum, University of Tartu, Tartu, Vanemuise 46, Estonia, loodusmuuseum@ut.ee

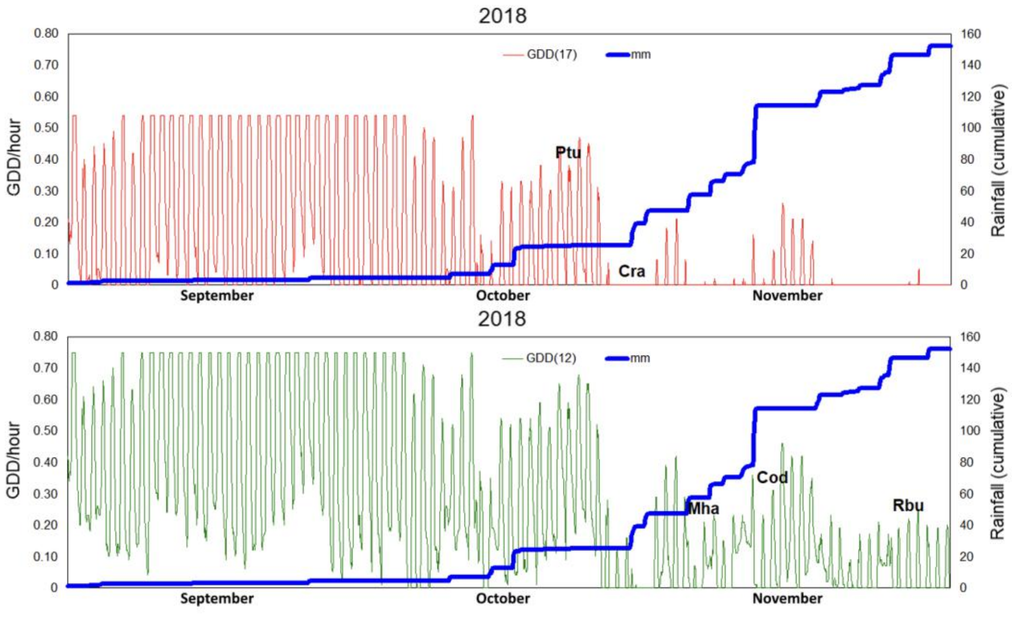
Red-listing is a globally acknowledged approach for assessing species' status to help practical addressing of the biodiversity crisis. The fungi are a highly diverse kingdom for which the species' conservation status remains poorly assessed due to taxonomic and detectability problems, but massive data are currently accumulating through molecular sampling techniques. We explored whether DNA metabarcoding data of soil fungi would change their red-listing assessments that are currently fruit-body based. We pre-selected sets of a total of 1,583 regionally assessed target species, and comparatively analysed both fruit-body and DNA barcoding records deposited in the public databases. We studied the correspondence of species' relative abundance and range parameters used in red-listing based on the two datasets. An average soil sample increased the range estimates of threatened and Near Threatened species by 0.18%. At 500 samples collected, their range estimates almost doubled and 45% previously unrecorded species were found. However, even after accumulating >1000 soil samples, about half of the fruit-body recorded species on the regional Red List remained undetected. The results indicate that one can distinguish certain fungal groups, based on their biological characters, where molecular datasets could profoundly change red-list assessments. For molecular data to become useful for fungal conservation, the crucial step is publishing the existing datasets and testing different approaches to how these can be incorporated in the red-listing protocols.

Predictability of Fruiting as a Function ofGrowing Degree-Days.

Paulo Oliveira[1,2], Carlos Vila-Viçosa[1,3]

[1]BIOPOLIS Program in Genomics, Biodiversity and Land Planning & CIBIO, 4485-661 Vairão, Portugal, [2]MED/University of Évora, Apartado 94, 7002-554 Évora, Portugal, oliveira@uevora.pt, [3]Herbário PO, Museu de História Natural e da Ciência da Universidade do Porto, 4099-002 Porto, cmvvicosa@mnhnc.up.pt, Portugal

Fruiting of macrofungi depends on intrinsic (autoecological) as well as extrinsic conditions, foremost among the latter are temperature and water availability. For Mediterranean ecosystems in particular, the attainment of these extrinsic conditions is difficult to predict each year, and tools to assist in translating them into predictions of fruiting are still to be developed. In this study we provide evidence that GDD (Growing Degree-Days), a common tool for predicting seasonal events in agriculture crops, such as flowering or pests (1), can provide a basis for such translation. Specifically for the Fall-Winter fruiting season, we have hypothesised that the decline in GDD, together with water availability, triggers fruiting.A relic *Quercus faginea* forest patch (2) was monitored for macrofungi fruiting during the Fall-Winter seasons between 2014 and 2021-2, and a retrospective analysis of the weather conditions was made in order to work out causality clues from the GDD and rainfall measurements. No weather stations were available in the vicinity, but the simulated data from meteoblue (history+ module) provided a reliable approximation (3). A good match to the phenological variations between years was obtained using T_{base} = 17 ºC for "thermophilic" species, and T_{base} = 12 ºC for the remainder species. We also analyse the GDD values for representative species with different intraseasonal timings and occurring over several years in this habitat, highlighting the potential of this approach (Figure 1). It will be argued that the principle of using GDD values, measured or simulated, is more broadly applicable, at least in temperate ecosystems, and that it can provide useful insights regarding climate change



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Preliminary investigation on antioxidant, antibacterial and antiproliferative activities on *Pleurotus eryngii* var. *thapsiae*, as potencial source of bioactive compounds.

Fortunato Cirlincione[1], Rossana V. C. Cardoso[2,3], Filipa Mandim[2,3], Adriana K. Molina[2,3], Tânia C. S. P. Pires[2,3], Giuseppe Venturella[1], Lillian Barros[2,3]

[1] Department of Agricultural, Food and Forest Sciences, University of Palermo, Viale delle Scienze, Bldg. 5, I-90128 Palermo, Italy, fortunato.cirlincione@unipa.it
[2] Centro de Investigação de Montanha (CIMO), Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal,
[3] Laboratório Associado para a Sustentabilidade e Tecnologia em Regiões de Montanha (SusTEC), Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal

Since ancient times mushrooms have been appreciated as food, and in more recent years their medicinal properties have been increasingly exploited. Apart from their use as simple foods, mushrooms are being explored as functional ingredients in the development of novel foods, mushroom-based products, and food supplements. Several studies on medicinal mushrooms have highlighted their wide pharmacological activities, including antioxidant, antimicrobial, anti-inflammatory, and antitumor properties. These pharmacological effects can be attributed to the rich bioactive metabolites composition found in fungi, primarily in the mycelium and sporophores (1).

The most studied molecules are phenolic compounds, proteins, fatty acids, terpenes, terpenoids, steroids, and vitamins (2). Several reports have also been conducted on polysaccharides, which are the structural components of the mushroom cell wall and exhibited a strong biological activity (3). Heteroglycans, peptidoglycans, and polysaccharide-protein complexes are polysaccharides that contribute to functional properties that mushroom are known to have, but the most abundant and well-known compounds are α- and β-glucans.

Pleurotus (Fr.) P. Kumm. is one of the most widely taxonomic group and include different cultivated and economically important mushrooms. These mushrooms are widely used in human nutrition all over the world and are also recognized as medicinal mushrooms. The genus includes facultative saprotrophs or biotrophs which grow in association with plants of the Apiaceae family, the *P. eryngii* species complex is undoubtedly noteworthy (4).

In this study, we focused on the use of *P. eryngii* var. *thapsiae* Venturella, Zervakis & Saitta, whose basidiomes grow in association with *Thapsia garganica* L. and have been currently reported so far only in Sicily (Italy). The basidiomes growing on two different substrates were used, the first based on wheat straw and the second based on a mixture of wheat straw and *Aegilops spp*., a common wheat weed. Aqueous crude extracts produced by two different techniques, conventional low-temperature, and ultrasound-assisted extraction, were used. These extracts were tested by radical scavenging activity (DPPH), thiobarbituric acid reactive substances (TBARS) formation inhibition, oxidative haemolysis inhibition assay (OxHLIA), antimicrobial (food and clinical bacteria and fungi) and antiproliferative assays on different human tumor cell lines. Preliminary results showed interesting antimicrobial activities against several Gram-positive and Gram-negative bacteria, including multi-resistant strains, as well as antifungal activities against *Aspergillus brasiliensis*. Regarding antiproliferative potential, very interesting results were observed against gastric (AGS), colorectal (CaCo-2), breast (MCF7), and liver (NCI-H460) cancer cell lines. Further evaluation of the biological activities and chemical characterization of polysaccharides within the extracts is in progress.

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Recruiting microorganisms for soil restoration: bioremediation of a co-contaminated soil using fungal bioresources isolated from a decommissioned military site.

Roberto Giovannini[1], Veronica Spinelli[1], Andrea Ceci[1], Mariarosa Bellino[2], Simona Scaffidi[2], Nunzio Capozzi[2], Anna Maria Persiani[1].

[1] Department of Environmental Biology, Sapienza University of Rome, Piazzale Aldo Moro, 00151 Rome, Italy.
Email: roberto.giovannini@uniroma1.it; veronica.spinelli@uniroma1.it; andrea.ceci@uniroma1.it; annamaria.persiani@uniroma1.it
[2] Centro Tecnico Logistico Interforze NBC, Italian Army, Ministry of Defense, Italy. Email: mariarosa.bellino@esercito.difesa.it; simona_scaffidi@yahoo.it; dir@cetli.esercito.difesa.it

Military sites, both active and decommissioned, possess a set of unique conditions (1) that lead to their particular dual nature. Several decommissioned sites in some EU States, due to the occurrence of species and habitat of interest, have been included in Natura 2000 network (2); conversely, both warfare and war-related industrial activities have noticeable impacts on affected sites (3). These complex yet largely unstudied sites have the potential to be sources of fungal bioresources, with potential applications in bioremediation as nature-based solution (1,4). To explore this hypothesis, this study has investigated the microbial strains (fungi and bacteria) isolated from soil and a *Plantago lanceolata*'s rhizosphere in a decommissioned site for their soil detoxifying potential in co-contamination conditions. To select the most suitable strains among those isolated from the site, a microwell plate enrichment experiment (5) was performed. Fungal and bacterial strains were tested for their ability to utilize PAHs as the sole carbon source while exposed to Pb and Zn in Czapek-Dox Broth. Strains showing growth in the tested conditions, evaluated using spectrophotometric readings, were selected for the subsequent phases of the study. As multi-strain consortia are gaining recognition for their higher bioremediation efficacy (6), an array of in-vitro compatibility tests was performed on the selected strains to evaluate the best candidates for the formulation of soil strains' synthetic consortia (6,7). Specifically, strains from the same sampling plot were tested in fungi-fungi and fungi-bacteria co-culture assays on Potato Dextrose Agar. As the *Trichoderma* genus has been largely reported for successful application in bioremediation, an additional test involving the five *Trichoderma* isolates in the group of selected strains was performed to assess the compatibility of strains belonging to the same genus isolated from different plots. To test the soil detoxification abilities of the selected strains and consortia, a microcosm experiment was set up. The microcosm involved 4 chemical treatments (PAHs, PAHs and Pb, PAHs and Zn, PAHs with Pb and Zn), 10 biological treatments and an uninoculated control treatment. The concentrations of the added soil contaminants were chosen in relation to the contamination thresholds specified by the Italian law on contaminated sites. The detoxification efficiency was evaluated using a batch of ecotoxicological assays (8) in order to select the most suitable treatment for future up-scaling. The results of the enrichment test highlighted 13 fungal strains out of 101 tested isolates and 15 bacterial strains out of 186 as able to utilize PAHs as their sole carbon sources in the tested condition. Subsequently to the compatibility tests, 9 consortia, including the 5-*Trichoderma* consortium, were formulated while one strain did not show compatibility with the others. Ecotoxicological assays results revealed that one treatment, in particular, was found to be effective in reducing toxicity in the microcosm experiment.

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Replacing Sanger sequencing by Nanopore sequencing for barcoding mushrooms using *Mycena* as a case study.

Cathelyn Maartje[1], Gallone Brigida[1], Nuytinck Jorinde[1,2]

[1]Naturalis Biodiversity Center, Darwinweg 2, P.O. Box 9517, 2300 RA Leiden, The Netherlands, maartje.cathelyn@naturalis.nl; [2]Research group Mycology, Department of Biology, Ghent University, K.L. Ledeganckstraat 32, 9000 Ghent, Belgium, jorinde.nuytinck@naturalis.nl

DNA barcoding has been a powerful tool for rapid species identification and generating species hypotheses in Fungi. However, the primary barcode for Fungi, namely the internal transcribed spacer (ITS), has proven to be problematic in some groups. In *Mycena* (Pers.) Roussel, for example, a large genus with a global distribution, ITS is too divergent. ITS is difficult to align at the genus level and possibly less reliable for identifying and delimitating species. In the current study, we combine a rapid DNA extraction method with third-generation sequencing methods to generate full-length rRNA operon sequences to assess taxonomic assignment accuracy and phylogenetic resolution, using *Mycena* as a case study. First, we used a magnetic beads-based method to extract DNA from fruiting body tissue collected in CTAB. Next, we amplified the rRNA operon using a single primer pair and sequenced the resulting amplicons using Oxford Nanopore Technologies (ONT). We predict that the methods used in the current study will result in better species identification and delimitation. Since the rRNA operon contains both conserved and variable regions we also expect it to be more suitable to reconstruct phylogenetic relationships at the genus level for *Mycena*. ONT sequencing enables researchers to combine up to 24 sample plates (i.e., 2,304 samples) in a single run, therefore, this method is far more efficient and less expensive compared to Sanger sequencing. Given these (and other) benefits, results from the current study could also inspire other researchers to test the utility of full rRNA operon sequences generated by ONT for other groups of Fungi.

Soil fungal communities in threatened polar and alpine environments.

Laura Zucconi[1], Federica D'Alò[1,2], Giorgia Cavallini[1], Fabiana Canini[1]

[1]Department of Ecological and Biological Sciences, University of Tuscia, Largo dell'Università s.n.c., Viterbo, Italy, zucconi@unitus.it, giorgia.cavallini@unitus.it, canini.fabiana@unitus.it
[2]Institute of Research on Terrestrial Ecosystems, National Research Council, Viale Guglielmo Marconi, 2, 05010, Porano (TR), Italy. federica.dalo@iret.cnr.it

Polar and alpine environments are among the most threatened by climate change, making them crucial hotspots for studying the effects of environmental shifts on soil ecosystems, which are the primary carbon sinks in cold environments. Although plant migration at higher altitudes and latitudes and areal expansion have been widely documented for the alpine regions as a consequence of climate change, it is still unclear how these phenomena may affect the richness, abundance, and functioning of the associated microbial communities, in particular soil fungi, which play a key role in nutrient cycling. We conducted various studies in the Alps to get insight into how plant-associated soil ecosystems may respond to warming in terms of microbial diversity and metabolic activity. Using Open Top Chambers (OTC) and comparing microbial communities along an altitudinal gradient as a proxy of future shifts, it was possible to estimate that warming is able to change the enzyme activity and that the migration of vegetation upwards may increase rates of microbial enzyme activity, increasing turnover of the labile and readily accessible soil organic matter and promote the decomposition of recalcitrant biopolymers (1). These results were supported by metagenomic and metatranscriptomic approaches (2), suggesting that the upwards migration of vegetation may accelerate recalcitrant C decomposition correlated with a higher increase of fungal share, thereby decreasing total ecosystem C storage. However, warming using OTCs had no effects on the fungal communities composition and functioning. In this optic, an new experiment using a transplantation approach is ongoing. Some soil monoliths, containing native plants, have been transplanted at a lower altitude (1000 meters shift) to simulate the effect of warming. Other monoliths have been used for a microcosm experiment at the CNRS-Ecotron of Montferrier-sur-Lez to simulate potential climate change scenarios. Instead, in Antarctica climate change is expected to lead to the disappearance of unique highly adapted ecosystems. Here we implemented studies focusing on understanding the relationship between the diversity and composition of soil fungal communities and the properties of their natural environments, and to monitor possible shifts related to environmental changes. All experiments resulted in a strong correlation between the fungal communities and the abiotic environmental conditions (3,4). Given the fact that hypotheses of the origin of microbial propagules in the continent are still contrasting, due to the geographic isolation of the continent and the limited wind dispersal from other continents, we are performing experiments to clarify the effect of local biological hotspots such as endolithic communities, on the diversity of soils in the most remote areas. This is of extreme importance, due to the expected dramatic impacts of the introduction of cosmopolitan more competitive species that will be favorited by milder climate conditions, at the expense of native taxa.

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Soil fungal communities of *Ulmus laevis* grown at forest and non-forest habitats.

Marta Kujawska[1], Maria Rudawska[1], Tomasz Leski[1]

[1]Institute of Dendrology Polish Academy of Science, Parkowa Str. 5, 62-035 Kórnik, Poland,
mkowalska@man.poznan.pl

Elms represent an important group of riparian forest trees primarily occurred in the north temperate regions. Both their ornamental value and the adaptability to stressful conditions, makes elms favorites along roadside avenues and in urban environments. Dutch Elm Disease (DED), one of the most destructive diseases of woody trees ever known causes by the highly aggressive fungal pathogen *Ophiostoma novo-ulmi,* has inflicted heavy losses counted in millions of elm trees with the near-total disappearance of adult trees in many parts of the Europe, Asia and North America. Since 1910, when DED began in Europe, the available scientific data are indeed vast and covers many aspects. However, these data have focused largely on disease-related issues, and neglected other aspects of elm biology. The mycobiome plays a significant role in soil ecological processes. A special part of mycobiome is represented by mycorrhizal fungi. Mycorrhizal fungi, in concert with soil fungi of other trophic groups, colonize plant roots, providing benefits to the host by improving plant nutrition and health, which is particularly important for host plants affected by inflict destructive global disease. Because of the multifaceted involvement of fungi for the health, nutrition and productivity of plants grown in different environmental conditions, it is important to have a full knowledge of the factors that influence mycobiome. Understanding the forces that govern the assembly of mycobiomes accompanying elms is key to the sustainable management of these forgotten tree species. As an object of research, one of the three elm trees native to Poland – *Ulmus laevis* has been selected. This elm species is widely distributed in most parts of Poland and best survived after the DED. To determine the structure of soil fungal communities accompanying roots of *U. laevis* eighteen locations, representing both forest (riparian and oak-hornbeam forests) and non-forest (forest nurseries, urban, rural, and post-industrial sites) habitats were chosen. At each location, the same elm trees were sampled during the two sampling periods – spring and autumn 2021. Both the mean species richness and the Shannon diversity coefficient significantly differ between the studied habitat types. The highest mean species richness was found in oak-hornbeam forests, and the lowest in forest nurseries. Our results indicate that habitat type, but not season, significantly influenced the soil mycobiome structure of *U. laevis*. Arbuscular mycorrhiza colonization was higher in the forest than in non-forest habitats, which is particularly indicated by data from urban and post-industrial sites, where the share of non-mycorrhizal roots was over 50%. Metagenomics analysis also confirmed that forest habitats were characterized by a higher share of symbiotic fungi than non-forest habitats. In turn, the level of colonization of arbuscular fungi was not affected by the type of habitat. This work was supported by the National Science Centre, Poland [grant no. 2020/37/N/NZ9/01915].

Soil fungi as a tool to contrast the diffusion of invasive vegetation species in coastal sand dunes.

Fabiana Canini[1], Sara Magrini[1,2], Beatrice Di Maio[2], Daniele Vela [2], Laura Zucconi[1]

[1]Department of Ecological and Biological Sciences, Tuscia University, Largo dell'Università s.n.c. Viterbo, Italy, canini.fabiana@unitus.it, zucconi@unitus.it; [2]Tuscia Germplasm Bank, Tuscia University, Largo dell'Università s.n.c.,Viterbo, Italy, magrini@unitus.it, beatrice_dimaio@libero.it, danielevela14@gmail.com

Species of the genus *Carpobrotus*, initially introduced to stabilize coastal dunes or for ornamental purposes, have spread and naturalized along the coasts of both northern Europe and the Mediterranean basin, due to their high rates of vegetative propagation and sexual reproduction (1), causing the rapid disappearance of native plants and the associated biodiversity. In particular, *Carpobrotus* litter exerts a short- to medium- term allelopathic effect on native species, reducing their seeds germination and establishment rates (2). Regardless, the impact of the *Carpobrotus* diffusion, its allelopathic effect on soil communities, and the potential role of microbial communities in its invasiveness in such limiting environments have not been clearly determined. Additionally, due to their wide metabolic competencies, fungal strains native to *Carpobrotus*-invaded soils may have evolved the ability to cope with the toxic compounds released by the plant and may be targeted as a tool for contributing to the revegetation of invaded areas. In this regard, soil samples have been collected in three localities of central Italy, in association with different species, such as *Anthemis maritima*, *Cakile maritima*, *Centaurea sphaerocephala*, and *Pancratium maritimum*, selected in both natural and *Carpobrotus* invaded areas. Fungal strains have been isolated from all the samples and the most frequent ones have been selected to test their resistance to the *Carpobrotus* allelopathic effect, in terms of ability to grow in the presence of water extracts from leaves of *C. edulis* and *C. acinaciformis*. Some of the strains tested showed enhanced growth in the presence of the extracts. Given that, we set up some tests on different plant species native to Italian coastal dunes: *A. maritima*, *Matthiola sinuata*, *Medicago marina*, *C. spherocephala*, *Malcolmia littorea*, *Plantago maritima*. We assessed their ability to germinate and grow in the presence of the *Carpobrotus* extracts, as well as when three of the strains most resistant to *Carpobrotus* were inoculated with the extracts, singularly and in combination, into the soil. We found a strong decrease in the growth of the plantlets in the presence of both *C. edulis* and *C. acinaciformis* extracts, but we observed an enhanced growth when fungal inocula were added to the extracts. Additionally, total soil fungal diversity has been characterized by a DNA metabarcoding approach; it has been related to the composition of the aboveground vegetation, the presence or absence of *Carpobrotus* species, and several soil physicochemical parameters. We revealed a strong variability of physicochemical parameters in the three localities that was connected to a strong variability in the communities diversity and composition. This did not allow to identify clear patterns connected to the *Carpobrotus* invasion common to all localities, even if some changes in the fungal communities were found for the single localities and for the single native plant species.

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Solidago canadensis and fungi. How plant invasion influence mycocoenoses.

Marta Wrzosek[1], Kamil Kisło[1], Marcin K. Dyderski[3], Barbara Kusińska[2], Marcin Mazurkiewicz[1], Katarzyna Rawlik[3], Agnieszka Adamska[5], Łukasz Kuberski[4], Patryk Czortek[2]

[1] University of Warsaw, Botanic Garden, Warsaw, Poland, ma.wrzosek@uw.edu.pl, [2]Białowieża Geobotanical Station, Białowieża, Poland, [3]Institute of Dendrology, Polish Academy of Sciences, Kórnik, Poland, [4.] Forest Research Institute, Białowieża, Poland, [5.] Inst. of Environmental Biology, Faculty of Biology, University of

The impact of alien species on ecosystems is intensively studied worldwide. Methods for preventing damage caused by particularly undesirable plant and animal species are being sought. The influence of invasions on fungi is usually overlooked. Canadian goldenrod (*Solidago canadensis*) was introduced to Europe as an ornamental plant in the 18th century. It is now considered a highly invasive species in Europe and dominates unused land throughout Poland, with a tendency to encroach on forest ecosystems (1,2). It is a plant that strongly alters the environment by shading the soil surface, forming monocultural populations, due to its allelopathic properties and intensive rhizomes growth (3). Research on the impact of *Solidago* on other plants has been conducted in Poland since 2005 (4). In 2022, studies were initiated to understand the influence of Canadian goldenrod on fungi in the ecosystem. Aeromycological studies were conducted in August 2022, and *Solidago* leaves decomposition on meadow ecosystems adjacent to the Białowieża Forest in Poland was analyzed during the winter season of 2022/2023. The common occurrence of fungi from the genera *Arthrinium, Fusarium, Epicoccum, Cladosporium, Cercospora*, *Drechslera*, *Phaeospheria* and *Alternaria* was demonstrated in the air collected from flowering populations of goldenrod. It was shown that both the stems and leaves of goldenrod are covered by dark-septate fungi during the season, which do not have a high cellulolytic potential and rather delay the decomposition of the stems than accelerate it. *Apiosporium* in its asexual form *Arthrinium* were found on the stems, while widespread occurrence of *Septoria fumosa,* which released spores only in May of the following season, was observed on the decaying leaves. The decomposition rate of leaf biomass was investigated, indicating slow mass loss, and in some cases, even an increase in mass during the winter season. Successional changes in decomposing leaves were analyzed. The study combined in vivo and in vitro research methods, allowing the construction of ecological dependency networks influencing the decomposition rate of Solidago canadensis biomass, with a focus on the importance of Oomycota, nematodes, mites, tardigrades, and ciliates, as well as their interactions with fungi. The composition of organisms detected in probes from plots with different *Solidago* coverage sheds light on the influence of that plant on soil ecosystem functioning.

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Talaromyces spp. - the postharvest fruit pathogens in Serbia

Stefan Stošić[1], Svetlana Živković[1]

Institute for Plant Protection and Environment, Teodora Drajzera 9, 11000 Belgrade, Serbia, stefan.stosic@gmail.com;

The genus *Talaromyces* encompasses a wide range of species that are ubiquitous in nature, thriving in diverse habitats such as soil, plant debris, and indoor environments. They inhabit different climatic zones across the globe and play crucial roles in natural ecosystems, through participation in organic matter decomposition and nutrient cycling. Members of this genus are also important enzyme, pigment and mycotoxin producers, biocontrol agents, human pathogens and food contaminants (1). *Talaromyces* spp. can be plant pathogens and few studies pointed that out (2-5). A polyphasic approach was employed to identify isolates of *Talaromyces* originating from diseased stored fruits in our country and subsequently determine their plant pathogenicity. Following protocols recommended by Yilmaz et al. (1), morphological examinations were carried out on Czapek yeast autolysate agar (CYA), Malt extract agar (MEA) and Creatine sucrose agar (CREA). Mycelial growth was also tested at three incubation temperatures (5, 25 and 37°C) while micromorphological features were inspected from MEA-grown cultures. Internal transcribed spacer (ITS) and beta-tubulin (*BenA*) regions were used for multilocus sequence typing (1). Molecular sequences from our isolates were combined with the reference and verified sequences to construct a combined (ITS+*BenA*) maximum likelihood phylogenetic tree. A pathogenicity test was conducted on originating hosts. After combining the results from the identification procedures, two species of *Talaromyces* were isolated in this study - *Talaromyces minioluteus* from pear, quince, tomato and orange fruits, and *T. rugulosus* from pear and lemon fruits. Phenotypic characteristics were similar to the previous descriptions in the literature (1, 6, 7). In pathogenicity assays, all isolates were pathogenic on the originating hosts. The symptoms in artificial inoculations resembled those found in naturally infected fruits and the reisolates exhibited the same morphological traits as the original isolates. These are the first detections of *T. minioluteus* and *T. rugulosus* and the first confirmations of their pathogenicity on the beforementioned plant hosts. The research presented here sheds light on the previously known fungi that can also act as plant pathogens/food spoilage agents. Their presence on the fruit diminishes its quality and leads to economic damage. Since different fruit and vegetables are sometimes kept in the same storage facilities, these plant pathogens residing on them can also contribute to inoculum build-up and cross-contamination phenomena.

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The diversity of larger fungi in peatlands: ten years of plot-based monitoring complemented by barcoding and metabarcoding

Nina Filippova[1], Elena Zvyagina[1], Elena Rudykina[1]

[1]Yugra State University, 628012, Chekhova street, 16, Khanty-Mansiysk, Russian Federation, filippova.courlee.nina@gmail.com

Peatlands are widespread around the world, with several highly paludified regions, one of which is the Western Siberian plain. Fungi are an important part of the decomposer community of peatlands, playing a critical role in the aerobic decomposition in the upper peat layer. The research of fungal diversity of peatlands globally, started over a century ago with microbiological approach, later complemented by mycologists working by direct observation of fruitbodies of larger fungi, and recently intensified by molecular approach, including metabarcoding of environmental DNA (1). The community of larger fungi of the raised bog “Mukhrino” in Western Siberia was studied using plot-based monitoring throughout a 10 year observation period. The revealed species diversity was represented by approximately 500 specimens in the Fungarium of Yugra State University collection (YSU-F) (2). Selected specimens were used for barcoding of the ITS region to reveal a total of 95 species from 33 genera and three classes. The barcoding effort confirmed morphological identifications for most specimens as well as revealed cryptic species and several potentially new taxa. Based on regular all-season observations, we described the phenology of the community fruiting. The quantitative community structure based on fruiting revealed difference in abundance of species by 4 orders of magnitude, the rare species representing near half of the species list. The inter-annual fruiting abundance varied several times by the total number of accumulated fruitbodies per year. In order to complement the direct observations of fruitbodies by environmental DNA sequencing, four major substrates were subjected to metabarcoding analysis: peat, bog plants litter, wood and mycorrhizal roots. Totally 192 samples were extracted during field summer of 2022 in order of cover spatial and temporal community variability, substrate features and some methodological questions and were sequenced on Illumina MiSeq (length of reads – 300 bp on both sides fragments). The sequences were analyzed by the pipeline QIIME2. The resulted taxonomic composition of fungi and quantitative community structure revealed by direct observation and metabarcoding generally coincide, but both approaches add a proportion of unique species. To make the comparisons with global studies, we created an open access database of literature-based observations of fungi in peatlands based on about 120 previously published papers (comprising about 5000 records of about 1300 species) (3). As a result, the study created an accurate representation of taxonomic and quantitative structure of the community of larger fungi in raised bogs in the region based on combination of methodological approaches (long-term monitoring of fruitbodies, specimens sequencing and metabarcoding). The raw data of plot-based counts was published as a sampling-event dataset (4) and the sequenced specimens with the sequence information as an DNA-derived extension dataset in GBIF (5).

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The effect of bacteria spp. to stimulate *Tuber melanosporum* root colonization and growth of young seedlings of *Quercus ilex* L.

Bianca Ranocchi[1], Veronica Giorgi[2], Lucia Landi[2], Ivan Castelli[2], Gianfranco Romanazzi[2], Cristiano Peroni[3], Davide Neri[2], Alessandra Zambonelli[4], Antonella Amicucci[1]

[1] Department of Biomolecular Sciences, Urbino University, Urbino, Italy, b.ranocchi@campus.uniurb.it antonella.amicucci@uniurb.it
[2] Department of Agricultural, Food and Environmental Sciences, Polytechnic University of Marche, Ancona, Italy
[3] Agenzia per l'Innovazione nel Settore Agroalimentare e della Pesca "Marche Agricoltura Pesca", AMAP, Italy
[4] Department of Agricultural and Food Sciences, Bologna University, Bologna, Italy

Tuber melanosporum is an ectomycorrhizal fungus with economic importance (in addition to the ecological function) due to gastronomical value of its fruiting body. Historically truffles started to be cultivated in XIX century, in the meanwhile wild truffle production started to decrease and, starting from 1960s, improvements in agricultural techniques were studied and applied (Callot 1999) so that natural production in forests became a cultivated orchard where we cultivate the host plant in order to harvest the truffles. Nowadays, over 50% of the truffles harvested worldwide (Hall et al. 2003) are produced in orchards. Research is hence focused in improving truffle cultivation by understanding the interaction between plant and fungus and evaluating agricultural techniques able to maximize production. In this scenario, the quality of the seedlings and the quality of their inoculation is a strong discriminant for the success of planting for truffle production. Several studies investigated growth conditions promoting *T. melanosporum* proliferation in the field (Bonet 2006, Garcia-Montero 2014, Buntgen 2015); others have focused on the standardization of the procedures for the evaluation of seedling quality in defense of customers (Alvarado 2013, Murat 2015), others focused on management practices in the greenhouse stage (Pruett 2009, Bruhn 2013). In the present study, we report the experimentation carried out on *Quercus ilex* seedlings mycorrhized with *T. melanosporum* and inoculated with three different formulations of selected bacterial species: one with *Bradyrhizobium* spp., one with *Pseudomonas* spp. and anorther with both. The aim was to test their potential ability to improve the quality of seedlings and improve mycorrhizal symbiosis establishment.

T. melanosporum colonization was assessed using morphological and molecular methods. Shoot growth and root parameters were analyzed. Our investigation showed in the early stages of ectomycorrhizal symbiosis on roots, a higher ECM colonization in the proximal (closer to the collar) section of roots apparatus than in the distal one. In detail a major effect was observed on roots co-inoculated with *Pseudomonas* spp., increasing the truffle inoculation rates of root tips by more than 5% compared with seedlings that received non-bacterial inoculation treatments. The encouraging results lay the foundations for the use of biosustainable strategies to improve the mycorrhization of truffle-producing plants.

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The fungal plastisphere in marine and terrestrial environments: combining culture-dependent and culture-independent approaches.

Matteo Florio Furno[1], Federica Spina[1], Francesco Venice[1], Giulia Stilo[1], Davide Ferrero[1], Viktoria Ilieva[2], Anna Poli[1], Valeria Prigione[1], Anita Fiorin[1], Giovanni Di Benedetto[2], Maya Dodhia[3], Nicole R. Posth[3], Pierangiola Bracco[2], Marco Zanetti[2] and Giovanna Cristina Varese[1]

[1]University of Torino, Dept of Life Sciences and Sysatems Biology, Viale Mattioli, 25 – 10125 – Torino, Italy; [2] University of Torino, Dept of Chemistry, via Giuria, 7 – 120125 -Torino, Italy; 3 Univerisity of Copenhagen, Dep of Geosciences and Natural Resource Management, Nørregade 10, PO Box 2177, Copenhagen, Danemark.

Plastic pollution is an environmental issue in both aquatic and terrestrial ecosystems. In 2013, the term "plastisphere" was coined by Zettler et al.[1] to refer to this new human-made ecosystem where plastic debris provides a substrate that can be colonized by microorganisms through the formation of a biofilms. The study of microbial biodiversity of the plastisphere and its metabolome is therefore fundamental for the development of several biotechnological applications. Nowadays, several microorganisms are known to be promising plastic degraders, fungi included[2]. This work aims to analyze the mycobiota associated with the plastisphere in both marine and terrestrial ecosystems. Marine fungi associated to different plastic polymers have been studied by means of culture dependent and independent approaches providing a wide understanding of the microbial community changes in response to anthropic pressure. Both water and sediments were considered, with sampling campaigns that involved Tyrrhenian Sea in Italy and Nordic Sea in Denmark. In the first case study, differently anthropized environments (e.g. the harbor of Livorno, a marine protected area and an intermediate point) were evaluated. When different kind of plastic polymers were used, the mycobiota did change indicating how fungal plastisphere is indeed adapted to each peculiar ecological niche. As regard terrestrial ecosystems, several fungi were isolated from a plastic-polluted landfill soil and their degradation abilities against conventional and biodegradable polymers were explored, isolating hundreds of strains. Few of them have been characterized for the capability to transform persistent (PE) and biodegradable plastics (e.g., PBS). Results showed that fungi may act against these polymers producing significant superficial modification of PE but led to an almost complete disintegration of the bioplastic. The degradation was confirmed by liquid trials where the produced transformation monomers were followed. This unexplored fungal biodiversity will be useful to face this emerging environmental problem, targeting both conventional plastics and biodegradable polymers in order to decrease the impact of plastic discharged in different ecosystems. Moreover, the enzymatic pathway involved in the degradation process would be useful to build up innovative enzymatic treatments for the waste management to allow a plastic circular economy. Finally, expanding the knowledge on plastisphere microorganisms and on their bioremediation capability would help tailoring new macromolecules accessible for microbial degradation.

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The threat posed by the invasive fungal pathogen of pines *Heterobasidion irregulare* in Europe: observations and predictions

Paolo Gonthier[1], Matteo Garbelotto[2]

[1]University of Turin, Dept. of Agricultural, Forest and Food Sciences, L.go P. Braccini 2, 10095 Grugliasco, Italy, E-paolo.gonthier@unito.it; [2]University of California at Berkeley, Dept. Environmental Sciences Policy and Management, Mulford Hall, Berkeley, CA 94720, USA, E-mail: matteog@berkeley.edu

Heterobasidion irregulare is a destructive fungal pathogen of pines native to North America and currently recommended for regulation by EPPO. This basidiomycete was introduced during WWII in central Italy, where the native congener *H. annosum* is also present. In this talk we review the findings that could help to predict the future trajectory of this alien fungal plant pathogen in Italy and in the European continent. While *H. irregulare* showed a greater sporulation potential, a faster growth rate in wood and a broader host range than the native *H. annosum*, precise predictions of future impacts of *H. irregulare* have been hampered by both the high fragmentation of conifer stands and the low incidence of *H. annosum* in central Italy. Population genetics studies allowed to predict that as the fragmentation of conifer stands decreases, the spread rate of *H. irregulare* will increase. Unexpectedly, experiments showed that the presence of the native *H. annosum* may accelerate and not slow down the spread of *H. irregulare*. Both results paint a troublesome scenario for areas in central and northern Europe characterized by large contiguous forest stands infested by *H. annosum*. Are these predicted scenarios likely to be true? In order to answer this question, we have studied the interaction between the two pathogens in central Italy. Field evidence shows not only that *H. irregulare* is truly invasive with a spread rate of 139 ha/year in a contiguous forest, but also that it is replacing *H. annosum* (1). Unexpectedly, part of the genome of *H. annosum* is being replaced by genes from *H. irregulare*, thanks to gene introgression mediated by interspecific hybridization. In addition, *H. annosum* genotypes containing *H. irregulare* genes increase their transmission-traits to levels comparable to those of *H. irregulare* (2). We conclude that Europe is facing two invasions: one by *H. irregulare* individuals and one by *H. annosum* individuals modified by the acquisition of *H. irregulare* genes. Both invasions are associated with disease that will spread at a faster rate than that caused by native *H. annosum*, thus increasing the estimated 800 million Euros per year of damage already caused by native *Heterobasidion* spp. Current ongoing experiments conducted in the framework of the Agritech National Research Center funded by 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) aimed at assessing the combined effects of rising temperatures and infections by *Heterobasidion* spp. on key conifer species will allow to refine the predictions on spread and impact of *H. irregulare* under climate change scenarios.

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Unraveling the complexity of antibiotic-related genetic machinery in pan-European soil metagenomes

Olesya Dulya[1], Vladimir Mikryukov[2], Alberto Orgiazzi[3], Leho Tedersoo[4]

[1]Chair of Mycology, Institute of Ecology and Earth Sciences, University of Tartu, J. Liivi 2, Tartu 50409, Estonia, olesia.dulia@ut.ee; [2]Chair of Mycology, Institute of Ecology and Earth Sciences, University of Tartu, J. Liivi 2, Tartu 50409, Estonia, vladimir.mikryukov@ut.ee; [3]European Commission, Joint Research Centre Ispra (JRC Ispra), Via Enrico Fermi 2749, 21027 Ispra, Italy (VA), alberto.orgiazzi@gmail.com; [4]Mycology and Microbiology Center (MMC), University of Tartu, J. Liivi 2, Tartu 50409, Estonia, leho.tedersoo@ut.ee

The global threat of antibiotic resistance and demand for advanced drugs motivate the exploration of microbial antibiotic biosynthesis and resistance in human-altered environments as well as undisturbed habitats. To understand environmental drivers and land use effects on the diversity of soil biota and their functional potential, we performed metagenomic sequencing of soil samples collected across Europe and provided with habitat and edaphic characteristics. In the analysis of metagenomes, we focused on gene modules responsible for antibiotic synthesis and resistance. Of 670 searched gene functional orthologs (hereafter genes) involved in the biosynthesis of secondary metabolites with antibiotic activity, we found 241 prokaryotic and fungal genes. Ninety-nine of read counts of the antibiotic biosynthesis genes belonged to Prokaryota, with Actinobacteria accounting for almost half of the reads, Gamma-, Beta-, and Deltaproteobacteria, Chloroflexi, Acidobacteria, Firmicutes, Cyanobacteria, and Bacteroidetes for two–five percent. Fungi possessed 0.14% of all reads, accounting for up to 20% of the abundance of three genes responsible for synthesizing enediynes and isopenicillin N, and for the total abundance of the genes encoding fumitremorgin, aflatoxin, meleagrin, and neoxaline production. These results imply that fungal genetic machinery for antibiotic resistance is substantially underexplored compared to bacterial and especially actinobacterial. In total 485 genes conferring antibiotic resistance were identified with 177 genes encoding drug efflux pumps.

Soil pH, land cover type, climate temperature and humidity were the main predictors of the relative abundance of both groups of genes. While the genes responsible for antibiotic biosynthesis and evolutionary ancient and taxon-specific drug efflux pumps were strongly related to environmental parameters and land cover type, evolutionary novel antibiotic resistance genes were more abundant in croplands, most likely due to increased proportion of bacterial DNA in communities.

The most interesting result of the work suggests that the distribution of the genetic machinery responsible for synthesizing a particular antibiotic across habitats and environmental gradients can be driven by the distribution of taxa synthesizing the antibiotic and also taxa which are targeted by the antibiotic. For example, in accordance with the dominance of fungi in forest soils (1), the genes responsible for the production of antifungal fumitremorgin and aflatoxin (synthesized exclusively by fungi) are much more relatively abundant in forests and strongly negatively related to soil pH. Also, gene families intrinsic to various bacterial taxa, such as Beta-, Gamma-, and Deltaproteobacteria, Actinobacteria, Cyanobacteria, and Bacilli (2) but providing the production of primarily antifungal kanosamine and pyrrolnitrin also have a higher relative abundance in forests and acidic soils. The abundance of bacterial gene clusters responsible for the production of various antibacterials are higher in cropland soil and soil with higher pH.

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Unrevealing the Hidden Secrets of *Hericium erinaceus* Mycelium and Sporophore trough Comparative Proteomics

Lorenzo Goppa[1], Carlo Santambrogio[2], Emanuele Ferrari[3], Stefania Brocca[2], Erika Ponzini[4], Rebecca Michela Baiguera[1], Paola Rossi[5], Elena Savino[1]

[1]Department of Earth and Environmental Sciences, University of Pavia, via Sant'Epifanio 14, 27100 Pavia, Italy, lorenzo.goppa01@universitadipavia.it, rebeccamichela.baiguera01@unipv.it, elena.savino@unipv.it
[2]Department of Biotechnology and Biosciences, University of Milano-Bicocca, Piazza della Scienza 2, 20126 Milan, Italy, carlo.santambrogio@unimib.it, stefania.brocca@unimib.it
[3]Institute of Biomedical Technologies, National Research Council of Italy, Via Cervi, 93, 20090 Segrate (MI), Italy, emanuele.ferrari@cnr.it
[4]Department of Materials Science, University of Milano-Bicocca, Via R. Cozzi 55, 20125 Milan, Italy, erika.ponzini@unimib.it
[5]Department of Biology and Biotechnology "L. Spallanzani", University of Pavia, via Ferrata 1, 27100 Pavia, Italy, paola.rossi@unipv.it

Proteomics is the identification and quantification, performed in a particular moment, of the complete set of protein in a biological sample. Recently, proteomics analysis has been applied also to mushroom sample as a powerful tool for investigating the proteome composition and functional dynamics of fungi.

These kinds of analysis enable to unravel the intricate protein networks underlying fungal biology and sheds light on their adaptation strategies and responses to environmental cues.

Although it is an extremely promising technique, nowadays fungal proteomics has been applied only to a few species.

Thanks to its widely demonstrated medicinal properties, among mushrooms of greatest interest to humans, *Hericium erinaceus* (Bull.) Pers. certainly stands out. However, studies demonstrating medicinal properties of *H. erinaceus* frequently do not precisely identify which molecule(s) are responsible for observed effects.

To address these needs, the present study aims to describe the proteome of *H. erinaceus* in both its growth phases: mycelium and fruiting body, highlighting peculiarities and potentiality of each.

For mycelium preparation and sporophores cultivation, the strain He2 from the MicUNIPV collection was employed.

Mycelium was grown up in a liquid culture of Malt Extract 2% and after 30 days collected and lyophilized. The dried sporophores were grown previously at the Botanical Garden of University of Pavia (Italy) on a substrate composed by 70% of oak sawdust, 20% of rice bran, and 10% wheat straw.

Then, 400 mg of each sample have been plunged in liquid nitrogen, reduced in powder and 100 mg of the resulting material has been suspended in 1 mL of Tris buffer in the presence of glass beads, homogenized, and centrifuged to separate soluble and insoluble fractions (SF and IF, respectively). IF has been suspended in Tris buffer and urea, and centrifuged to remove beads. Proteins in SF and IF samples have been reduced, alkylated and finally digested by trypsin.

Samples have been analyzed by LC/MS employing an Orbitrap Fusion instrument equipped with a HPLC system; proteins identification and quantitation has been performed by Proteome.

Analysis allowed the identification, on average, of 1588 different proteins for the mycelium and 1435 for the sporophore. Among these, 360 were unique to the mycelium and 503 to the sporophore. Subsequent statistical analysis, performed using Perseus software, enabled the identification of 70 differentially expressed proteins between the two samples: 23 in the mycelium and 47 in the sporophore.

Through Gene Ontology analysis (Biological Process), it was found that in the mycelium proteins involved in steroid biosynthetic process, regulation of DNA replication, protein folding, methylation, and biosynthetic process were overexpressed.

On the other hand, proteins involved in response to oxidative stress, polysaccharide catabolic process, potassium ion transport, and phosphatidylethanolamine biosynthetic process were overexpressed in the sporophore.

Untangling diversity and adaptation of rock-inhabiting fungi in drylands

Claudia Coleine[1], Laura Selbmann[2]

[1]Department of Ecological and Biological Sciences, University of Tuscia, Largo dell'Università, snc, Viterbo, 01100 Italy Institution, coleine@unitus.it; selbmann@unitus.it

Endolithic communities are microbial ecosystems dwelling inside porous rocks that are able to persist in arid regions worldwide up to the edge of the biological potential for life. In hyper-arid environments, once the threshold of dry tolerability for plants and soil microbes is crossed, endoliths represent the ultimate life-forms, regulating water retention and creating positive feedback for ecological successions. Rock-dwelling fungi, in particular, play critical ecological roles in drylands, including nutrient cycling; however, we know very little about the diversity, function and environmental preferences of these important organisms, and even the amplitude and distribution of rock mycobiome across diverse arid regions of the planet still remains undetermined. Endolithic communities dominated by lichenized fungi in Antarctica represent to date the most well-know and studied rock communities; they colonize the ice-free areas of the continent, including the McMurdo Dry Valleys, often accounted as the closest terrestrial counterpart of the Martian environment and thought to be devoid of life until the discovery of these cryptic life-forms. Despite their interest as a model for the early colonization of terrestrial ecosystems, little is known about the evolution, diversity, and genetic makeup of microbial species that reside in these environments and about the importance of inter-kingdom interactions that improve the community fitness. To address this knowledge gap, we generated the first metagenomes from 109 rocks collected in Continental Antarctica over a distance of about 450 km along an altitudinal transect from 834 up to 3100 m above sea level (a.s.l.). We generated 4,539 metagenome-assembled bacteria genomes, 49.3% of which were novel candidate species and presented evidence that trace gas oxidation and atmospheric chemosynthesis may be the prevalent strategies supporting metabolic activity. We also retrieved a viral catalog comprising > 75,000 viral operational taxonomic units and predicted auxiliary metabolic genes (AMGs) with functions indicating that they may be potentially influencing bacterial adaptation and biogeochemistry. Finally, we assembled 266 eukaryotic genomes, of which 230 were fungi dominated by Ascomycota, mostly representing new fungal candidate species and Tremellomycetes (Basidiomycota). To date, this resource represents the largest effort to capture the breadth of microbial genomic diversity from Antarctic rocks, considerably expanding our perception of the phylogenetic diversity of inhabiting taxa, focusing on key tolerance competences that promote their spreading and perpetuation at the fringe for life sustainability. Finally, with the European Union funded project DRYLIFE, we have also recently extended our research to endolithic communities from hot deserts, i.e. US Western drylands, which represent 40% of land in the US and where a 3°C temperature increase is expected by the end of the century. The results from this project may optimize future climate impact projection on drylands and deliver information that policy makers need.

What lies beneath the logs: a collection of polyporoid and corticoid strains of central and northern Spain.

Simone Buratti[1], Sergio Pérez Gorjón[2], Carolina Elena Girometta[1], Elena Savino[1].

[1]University of Pavia, Department of Earth and Environmental Sciences, via Ferrata 1, 27100, Pavia, Italy, simone.buratti01@universitadipavia.it; carolinaelena.girometta@unipv.it; elena.savino@unipv.it
[2]University of Salamanca, Department of Botanic and plant physiology, Biobanco de ADN Vegetal, Facultad de Biología, Calle Licenciado Méndez Nieto s/n, 37007, Salamanca, Spain, spgorjon@usal.es

In forests ecosystems, polyporoid and corticioid fungi are the most represented and common wood inhabiting organisms. These two groups have important roles for many of the natural dynamics in which they are involved, but also represent a source of great value for the scientific world. Every year scientific research brings to light discoveries and applications related to the use of lignicolous fungi in fields such as medicine, nutraceutics and biotechnology. Polyporoid fungi are a well-studied group when compared with corticioid fungi, about which relatively little is known about their potential applications. The value of fungi rises with the birth of mycological collections, which collect, preserve, and study strains and species that may have unexplored potential. Collections are also important to ensure exchange and availability of quality-guaranteed and diverse strains at international level. The aim of this study was the collection and the identification of corticioid and polyporoid fungi from two biogeographic areas of Spain: the Mediterranean zone from the center of Spain and the Eurosiberian oceanic climate zone from the north of the Iberian Peninsula. All 128 basidiomata collected were identified morphologically and, wherever possible, isolation of mycelium in pure culture was performed from spores or directly from basidiomata. The mycelium of 54 strains was successfully isolated and further confirmed by ITS-based molecular identification of DNA. These strains represent the first collection of lignicolous fungal strains of the University of Salamanca, that are now deposited in the SALA-DNA Plant Biobank. Growth rate of each isolated strain was tested and mycelium characteristics such as color, colony morphology and microscopic features were recorded. These characteristics are an important starting point in order to select potentially useful and interesting species for future applications where even simple characteristics such as texture, speed, and color can become reasons for strong strain selection. Among isolated species notable entries for taxonomical studies are: *Botryobasidium asperulum* since its presence in Europe is controversial and no ITS sequences are available and *Phlebia rufa*, probably a species complex with several critical sibling entities for which more phylogenetic studies are needed. Some isolated species have also already well-established potentials such as: *Hericium erinaceus*, *Grifola frondosa*, *Pleurotus pulmonarius*, *Pleurotus eryngii* var. *ferulae* and *Fomitopsis pinicola* for their medicinal properties (1), *Schizophyllum commune* and *Trametes versicolor* for their use in medicinal and myco-remediation fields (2), *Trametes hirsuta* and *Stereum hirsutum* respectively for myco-remediation and myco-materials (3). This study provides a valuable resource of strains to continue to deepen knowledge of already well-known species and to discover more about those less studied.

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What more is there to know about biology of morels (*Morchella* spp.) to improve their cultivation?

Mirco Iotti[1], Yue Huang[2], Carmelo Gianchino[1], Marco Leonardi[1], Alessandra Zambonelli[2]

[1] Department of Life, Health and Environmental Sciences, University of L'Aquila, via Vetoio, Coppito 1, 67100 L'Aquila, Italy, mirco.iotti@univaq.it, carmelo.gianchino@graduate.univaq.it, marco.leonardi@univaq.it;
[2] Department of Agricultural and Food Sciences, University of Bologna, via Fanin 44, 40127 Bologna, Italy, yue.huang2@unibo.it, alessandr.zambonelli@unibo.it

Morels (*Morchella* spp.) are edible mushrooms appreciated as gourmet food worldwide. The genus contains more than 80 phylo-species sorted into 2 main sections - *Morchella* (yellow morels) and *Distantes* (black morels) - and the basal section *Rufobrunnea* with only two species (1). The first successful attempt to produce morel ascomata in controlled conditions dates back to the early 1980s (2) but large-scale cultivation of morels has not spread in the following decades due to the low economic sustainability of the production process. Success of outdoor cultivation has been achieved only from 2011 with the introduction of exogenous nutrition bags (3) on the fruiting plots. In few years, China has become the world's leading producer and exporter of morels (4). Despite the rapid expansion of area under morel cultivation, cases of drops in production or unstable yield are frequent and cannot be explained by the current understanding of morel biology. The nutritional habit of *Morchella* spp. is highly variable and much still remain to be discovered (5). Species in the *Distantes* clade are saprotrophic and it is for this reason that *M. eximia*, *M. importuna* and *M. sextelata* are the unique cultivated species to date (4). In contrast, species in the *Morchella* clade establish different types of interactions (e.g., mycorrhiza-like and radiscisclerotia) with the roots of a wide range of host plants, the functioning of which is largely unknown. Also, different sexual reproductive modes have been detected in some species and both homokaryotic and heterokaryotic mycelia can be found in wild and cultivated strains (6). Another distinctive feature of *Morchella* spp. is the abundant fruiting in burnt forests, particularly the year following the fire event. Specific studies conducted on 4 different burnt areas in Italy showed that morel production was concentrated in woody patches where the fire intensity was moderate. The role of bacteria, particularly *Pseudomonas* spp., in the life cycle of morels is rather unknown but recent researches show that microorganisms can be efficiently used to improve the cultivation system by promoting the growth of mycelia and fruiting and increasing the tolerance to pathogens or environmental stresses (7). The improvement of morel cultivation technology must be accompanied by an in-depth screening and selection of wild genotypes for stress tolerance, proneness to fruit, growing substrates, etc. Once isolated and characterized new strains, protoplast fusion can then represent a promising bio technological approach to obtain new heterokaryotic stable genotypes overcoming the vegetative incompatibility barriers. In this perspective, a first attempt has been recently made to cultivate Italian strains of *M. importuna* by applying the same technology used to cultivate Chinese strains.

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Where are we in assessing the conservation status of European fungal species?

Gregory M. Mueller, James R.S. Westrip, Anders Dahlberg

[1]Chicago Botanic Garden, 1000 Lake Cook Rd, Glencoe, Illinois, 60022, USA and IUCN SSC Fungal Conservation Committee, Rue Mauverney 28, 1196 Gland, Switzerland, gmueller@chicagobotanic.org, ORCID:0000-0001-8316-9319; [2]Biodiversity Assessment & Knowledge Team, Centre for Science and Data, IUCN, The David Attenborough Building, Pembroke Street, Cambridge CB2 3QZ, UK. James.Westrip@iucn.org, ORCID:0000-0003-4467-080X, [3] Department of Forest Mycology and Plant Pathology, Swedish University of Agricultural Sciences, 75007 Uppsala, Sweden, anders.dahlberg@slu.se, ORCID:0000-0002-3669-6797.

Several recent papers have been published or are in press that use data included in the IUCN Red List of Threatened Species™ (henceforth Red List) to understand what is known about the conservation status of fungi at the global level (1, 2, 3,4). This presentation will review where we are in assessing the conservation status of European fungal species. While most European countries have at least some data on the conservation status of fungi at their national level (1, 4) the current update of the Red List (2023-2) (5) only includes 170 European species (143 Basidiomycetes and 27 Ascomycetes). Of these, 86 are of conservation concern, i.e., assessed as Critically Endangered, Endangered, or Vulnerable. The majority of these species are Basidiomycetes (74 of 86) with the other 12 species being Ascomycetes, 9 of which are Lecanoromycetes. Few patterns can be gleamed regarding the conservation status of European Ascomycetes with only 12 assessments currently published, so the focus of our synthesis is on the published Basidiomycete species. Currently, no published European Agaricomycetes species is assessed as globally Critically Endangered while 18 and 56 are assessed as globally Endangered and Vulnerable, respectively (5). With only 143 Agaricomycetes out of an estimate of at least 15,000 European macrofungi assessed, these data cannot be used to infer the true conservation status of European fungi. Additionally, the interpretation of these data is further challenged due to geographic and habitat biases in the selection of the species that have been assessed. However, the published data do provide a preliminary summary of the situation that can be used to inform future efforts.

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Who’s there? Retrieving genetic markers from public databases to improve fungal species delimitation and identification

*Angela Conti [*1], Roberto Scarponi[1], Debora Casagrande Pierantoni[1], Laura Corte [1,3], Vincent Robert[2] and Gianluigi Cardinali[1,3*]*

[1] Department of Pharmaceutical Sciences- University of Perugia – Italy; [2] Westerdjik Institute for Biodiversity – Utrecht Netherlands; [3] CEMIN Excellence Research Centre – Perugia Italy

ABSTRACT:

The assessment of fungal biodiversity is currently carried out either by analysis of single specimens or by metagenomics. In the former case, some fungal groups present objective problems to obtain DNA of sufficient quality for SANGER sequencing of the taxonomic markers. For all other fungi, and particularly for yeasts and yeast-like organism, ITS has long been established as the gold standard for the identification at the species level [1]. Similarly, this marker is largely employed as the sole locus in metagenomic studies to asses presence and frequency of the various fungal species, in few instances the older marker LSU D1/D2, aka 26S, is analyzed in addition to the ITS [2]. These sequences are located in the Ribosomal DNA operon characterized by several tens or even hundreds of copies [3]. In spite of an active concerted evolution, not all tandem repeats within the same genome are equal, thus producing a sort of background noise scarcely detected by SANGER, but quite evident as variants Next Generation Sequencing reads[4]. For this reason, single-copy protein encoding genes have been proposed as an alternative to the traditional “ribosomal markers” or as an additional source of data to combine for Multi Locus Sequence Typing (MLST). The main drawback of such genes is that they lack conserved regions for designing universal primers, and thus their amplification rate is low[1]. A genome-based approach could be employed to overcome this limitation. There are two main strategies that could be implemented: the first one consists in the retrieval of such genes directly from an assembled genome and the subsequent alignment against a database [5], while the second involves the direct mapping of raw reads obtained from whole genome sequencing against specific databases. In this scenario, there is the urgent need of robust, curated secondary barcodes databases [6]. Furthermore, this study aims at proposing MeTRe (Mean Taxonomic Resolution), a novel index able to evaluate both marker efficacy and the actual resolution (i.e. the level of separation) between species obtained with different markers or their combinations. Results shows that the use of four single copy markers (*TEF1*-α, *ACT1*, *RPB1* and *RPB2*) increases the resolution among the species.

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POSTER SESSION*

* Title sorted alphabetically according to the Congress program.

An impact of invasive trees species: black locust (*Robinia pseudoacacia*), black cherry (*Prunus serotina*) and northern red oak (*Quercus rubra*) on symbiotic soil fungi in native

Robin Wilgan[1], Marta Kujawska[1], Tomasz Leski[1]

[1]Institute of Dendrology Polish Academy of Science, Parkowa 5, 62-035 Kórnik, Poland,
rwilgan@man.poznan.pl

Biological invasions are a key element of modern nature conservation and management. Invasive tree species reduce biodiversity, modify nutrient cycles and turn native forests into novel ecosystems determined by invasive species. However, an impact of invasive tree species on ectomycorrhizal fungi in native forests is poorly studied. Ectomycorrhizal fungi establish a close symbiosis with tree roots and are crucial for the proper development of almost all European trees. This study is aimed to determine, how an invasion of *Robinia pseudoacacia*, *Prunus serotina*, and *Quercus rubra* affect symbiotic fungi in native forest ecosystems. In addition, other trophic groups of soil fungi were also analysed.
We used high-throughput sequencing (NGS sequencing) because this method allows to detection of thousands of fungal taxa based on DNA isolated from the environmental samples. We have taken soil samples along the gradient of increasing abundance of invasive tree species in native, pine-dominated forest ecosystems. We studied forest stands dominated by invasive trees, stands with a low abundance of invasive trees, and with no invasive tree species. Each invasive species was tested independently. Altogether, we studied 81 stands in Central and Western Poland.

The results have shown, that each of the three studied invasive tree species has a significant impact on soil fungi. A high abundance of *Robinia pseudoacacia* has the strongest negative effect leading to eradicating ectomycorrhizal fungi from soil. Low abundance of *Robinia pseudoacacia* and high abundance of *Prunus serotina* have similar and less negative effects on ectomycorrhizal fungi than *Robinia pseudoacacia*, while low abundance of both *Prunus serotina* and *Quercus rubra* has no significant impact on fungi. A high abundance of *Quercus rubra* leads to an increase in the share and diversity of ectomycorrhizal fungi compared to pine forests. Moreover, an increase in the abundance of each tested invasive tree leaded to a progressive decrease in the share of root-associated endophytic fungi and to the increase in the share of pathogenic fungi in soil.

Robinia pseudoacacia has also the strongest negative impact on the surrounding forest habitats. Pine forests with no invasive tree species located near *Robinia pseudoacacia* stands have significantly lower share and abundance of ectomycorrhizal fungi than pine forests elsewhere. Moreover, the pot experiment has shown a similar pattern. Native oaks and pines were seeded to soil taken from forest stands dominated by *Robinia pseudoacacia*, *Prunus serotina*, and *Quercus rubra*, and non-invaded pine forest. Two-years-old seedlings in soil from *Robinia pseudoacacia* dominated stands had significantly lower growth and biomass, than seedlings in other variants.

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Antifungal activity of *Asparagus officinalis* L. spear extracts against *Fusarium oxysporum* and *Fusarium proliferatum*

Monika Urbaniak [1*], Elsie Ayamoh Enow [1*], Sylwia Ryszczyńska [2], Agnieszka Waśkiewicz [2], Łukasz Stępień[1]

[1]Department of Pathogen Genetics and Plant Resistance, Institute of Plant Genetics, Polish Academy of Sciences, Strzeszyńska 34, 60-479 Poznań, Poland (E-mail: murb@igr.poznan.pl; eeno@igr.poznan.pl)
[2]Department of Chemistry, Poznań University of Life Sciences, Wojska Polskiego 75, 60-625 Poznań, Poland

Asparagus (*Asparagus officinalis* L.) is a very popular vegetable because of its exceptional taste and high nutritional value. This plant is often colonized by various pathogenic fungi belonging to the *Fusarium* genus, among which *Fusarium proliferatum* and *Fusarium oxysporum* are the most frequent species. These are well-known mycotoxin producers, mainly fumonisins FB_1-FB_3, beauvericin, enniatins, and moniliformin. *Fusarium* fungi might cause devastating plant diseases such as Fusarium wilt or Fusarium root rot, which limit asparagus production worldwide. On the other hand, asparagus plants induce a complex of mechanisms to adjust cellular functions and activate defense responses against *Fusarium* species. The plant attacked produces metabolites such as flavonoids that can affect *Fusarium* growth and mycotoxins production. However, detailed research is still needed to explain the basis of this plant-pathogen interaction. Therefore, the objective of this study was to assess the potential of *Asparagus officinalis* L. spear extracts obtained by supercritical fluid extraction to inhibit the growth of *F. oxysporum* and *F. proliferatum* strains and also to analyse the changes in the biosynthesis of mycotoxins in the culture medium under laboratory conditions. The experiment was conducted on PDA solid medium with different concentrations of plant extract (2.5, 5, 7.5, and 10%) applied to the culture of individual fungal strains, as compared to the control group (PDA with fungal strain without extract). Four fungal isolates (two *Fusarium oxysporum* and two *Fusarium proliferatum* isolates) were used as test pathogens. They were identified using molecular techniques and are maintained in the strain collection at the Institute of Plant Genetics, Polish Academy of Sciences, Poznań, Poland. Inoculated plates were incubated at 25°C in the darkness for 10 days, and the radial mycelial growth was measured every 3 days. For each treatment, three replicates were performed and the experiment was repeated three times. Finally, the antifungal activity of each extract was calculated in terms of the inhibition percentage of mycelia growth. The mycotoxin analyses were conducted by using UHPLC/MS/MS. The analyses were quantitatively analyzed using multiple reaction monitoring, and their identification was confirmed by comparing the retention times and m/z values with those of corresponding standards. The results have shown strong antifungal activity of *Asparagus officinalis* L. spear extracts against *Fusarium oxysporum* and *Fusarium proliferatum*. Similar results were obtained during our previous research using various plant extracts (*Lamium album*, *Sorbus aucuparia* or *Melissa officinalis*) prepared under the same laboratory conditions. Investigated plant extracts strongly affected *Fusarium* metabolism by inhibiting mycotoxin biosynthesis and/or mycelium growth.

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Bioactivity and metabolomic profile of extracts derived from mycelial solid cultures of *Hypsizygus marmoreus*.

Paola Angelini[1], Giancarlo Angeles Flores[1], Gaia Cusumano[1], Roberto Venanzoni[1], Roberto Maria Pellegrino[1], Carla Emiliani[1], Gokhan Zengin[2], Simonetta Cristina Di Simone[3], Luigi Menghini[3*], Claudio Ferrante[3]

[1] Department of Chemistry, Biology and Biotechnology, University of Perugia, 06122 Perugia, Italy; giancarlo.angelesflores@unich.it; gaia.cusumano@studenti.unipg.it; roberto.pellegrino@unipg.it; carla.emiliani@unipg.it; roberto.venanzoni@unipg.it
[2] Physiology and Biochemistry Research Laboratory, Department of Biology, Science Faculty, Selcuk University, 42130 Konya, Turkey; gokhanzengin@selcuk.edu.tr
[3] Botanic Garden "Giardino dei Semplici", Department of Pharmacy, "Gabriele d'Annunzio" University, Via dei Vestini 31, 66100 Chieti, Italy; simonetta.disimone@unich.it; luigi.menghini@unich.it; claudio.ferrante@unich.it

The beech mushroom (*Hypsizygus marmoreus*) is a highly nutritious, edible medicinal mushroom native to East Asia. It is also cultivated in temperate climates in Europe, North America, and Australia. The quality, nutritional value, and bioactive content of both mycelia and mushrooms are significantly influenced by the composition of the growth substrate. Hence, this study aimed to investigate the impact of different substrates on the metabolite compositions of *H. marmoreus* mycelia cultivated *in vitro* (1). The substrates tested included malt extract agar, malt extract agar enriched with barley malt, and malt extract agar enriched with grape pomace. In addition to analyzing metabolite compositions, the study also assessed the antimicrobial activity of the extracts against bacteria, yeasts, and dermatophytes. Broth microdilution method has been used to determine the antimicrobial activities and Minimum Inhibitory Concentrations (MIC) of different mycelia extracts against Gram-positive bacteria (*Bacillus subtilis* and *Staphylococcus aureus*), Gram-negative bacteria (*Escherichia coli*, *Salmonella typhi* and *Pseudomonas aeruginosa*), yeasts (*Candida albicans*, *C. tropicalis* and *C. parapsilosis*) and dermatophytes (*Trichophyton mentagrophytes*, *T. tonsurans, T. rubrum*, *Arthroderma quadrifidum*, *A. gypseum, A. curreyi* and *A. insingulare*).

The results revealed that the *H. marmoreus* mycelia extracts demonstrated both antibacterial and antifungal activities against the tested microorganisms. Extracts obtained from the cultivation in substrates enriched with either barley malt or grape pomace exhibited the highest antibacterial activity among all the tested bacterial strains, except for *P. aeruginosa*. On the other hand, the same extracts showed the highest inhibitory effect on *C. albicans* (YEPGA 6379) and *C. parapsilosis* (YEPGA 6551). Noteworthy, the extract from the mycelia cultivated in the substrate enriched with grape pomace also exhibited remarkable efficacy against *T. mentagrophytes* and *T. tonsurans* with MIC range of 6.25-12.5 µg µL^{-1} (GM: 9.92 µg µL^{-1}) and 12.5 – 25 µg µL^{-1} (GM: 19.84 µg µL^{-1}), respectively.

Moreover, antioxidant activity was assessed using DPPH and ABTS methods, and related to the content in total phenols and flavonoids. Finally, the mycelial extracts of *H. marmoreus* were also subjected to an investigation of their metabolic profile through untargeted LC/MS QTOF analysis. By examining these factors, we can gain valuable insights into how growth substrate selection affects the nutritional and medicinal properties of *H. marmoreus*, making it a valuable contribution to the understanding of cultivation of this mushroom.

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Biodiversity of ascomycetous fungi on forests in the Yenice district of Türkiye

Faruk Selcuk[1], Merve Ulukapı[1], and Hakan Allı[2]

[1]Kırsehir Ahi Evran University, Sciences and Arts Faculty, Department of Molecular Biology and Genetics. Kırşehir / TÜRKİYE, selcuk_faruk@yahoo.com, merveulukapi@gmail.com
[2]Muğla Sıtkı Koçman University, Sciences Faculty, Department of Biology. Muğla / TÜRKİYE, hakanalli@gmail.com

Biodiversity is a kind of a measure of variation at the genetic, species and ecosystem level which is not distributed evenly on Earth.Yenice Forest which is located in the West region of Karabük province, Türkiye with nine different biodiversity hotspots, is one of the significantly important strict preservation zones in Türkiye and has been declared as one of the hot spots of Europe for its rich biodiversity by World Wildlife Fund. Yenice has a cool summer and cold and rainy winter and the annual mean temperature of 13.6 °C. Since it belongs to the Western Black Sea climatic zone, the forest region experiences a humid, wet and misty climate throughout the year with an average annual precipitation of 6933 mm. Yenice composed mainly of pristine forests with needle-leaved and broad-leaved trees.

The significance of these Yenice forests is that its trees, shrubs, plants and wildlife exist together in higher numbers similar to tropical areas. According to recent studies, there are about 312 plant species residing in Yenice Forests including grand-bodied trees such as maple, yellow pine, black larch and sessile oak; medicinal herbs such as snowdrop, butcher’s broom, blueberry, mistletoe, and mullein; and monumental trees which reach diameters and heights rarely seen in the world forming independent ecosystems.

Microfungi are the most important causes of plant diseases. The detailed investigation of the mycota in natural or protected forest ecosystems and the dynamics and pathogen of mycobiota of plant populations in the areas exposed to anthropogen effect is an important step in struggle to the parasites and pathogen microfungi species in both phytocenosis and mycosinusia.

We conducted joint research on micro-macro Ascomycetous fungi during 2020-2023 in order to study and carry out the comparative analysis of ecology and biological diversity of them, in forest phytocenosis on the Yenice Forest.

During the project, field studies were carried out in the area and samples were collected from parts of trees and shrubs that form forests, such as branches, stem bark, leaves and fruits, which are infected with microfungi especially. As a result of the examination of the samples brought to the laboratory, 32 species were identified. According to the literature review, 4 genera and 10 species of Ascomycetous microfungi are new records for the Turkish mycobiota as a result of this research.

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Broadening the investigation area of *Tuber aestivum* genetic diversity.

Beatrice Belfiori[1], Claudia Riccioni[1], Eva Gómez Molina[2], Sergio Sánchez Durán[3], Sergi García Barreda[3], Žaklina Marjanovic[4], Fatima Henkrar[5], Lahsen Khabar[5], Luigi Gallo[6], Andrea Rubini[1]

[1]Istituto di Bioscienze e Biorisorse (IBBR), Consiglio Nazionale delle Ricerche (CNR), Via della Madonna Alta 130, 06128, Perugia, Italy, beatrice.belfiori@ibbr.cnr.it, claudia.riccioni@ibbr.cnr.it, andrea.rubini@ibbr.cnr.it; [2]Centro de Investigación y Experimentación en Truficultura, Partida Fabardo, s/n 22430 Graus (Huesca), España, egomez@dphuesca.es; [3]Centro de Investigación y Tecnología Agroalimentaria de Aragón (CITA), Avda. Montañana 930, 50059 Zaragoza, España, ssanchezd@cita-aragon.es, sgarciaba@cita-aragon.es; [4]University of Belgrade - Institute for Multidisciplinary ResearchBulevar despota Stefana 142, 11060 Belgrade, zaklina@yahoo.com; Université Mohammed V, Faculté des Sciences, Département de Biologie, [5]Centre de Recherche "Biotechnologies Végétale et Microbienne, Biodiversité et Environnement", Avenue Ibn Battouta B.P. 1014, Rabat, Maroc, f.henkrar@um5r.ac.ma, l.khabar@um5r.ac.ma, [6]Azienda Regionale per lo Sviluppo dell'Agricoltura Calabrese (ARSAC) - Ce.D.A. n.2 - Castrovillari (CS). luigi.gallo@arsac.calabria.it

Tuber aestivum is associated with a wide range of tree species and it is the most widespread edible truffle, being widely distributed not only in Europe, with populations ranging from Sweden to Spain, but also from Turkey to Middle East countries and North Africa (1, 2). Its capability of colonizing many different areas along with a long ripening period, has increased scientific studies and commercial interest in the last decades. A number of large- and fine-scale population genetics studies have been conducted to explore its genetic diversity and genetic structure. Recent investigations based on ITS sequence analyses revealed a strong phylogenetic pattern and a higher level of genetic diversity in Turkish and southern European populations than in northern European populations, suggesting that Turkey and southern Europe may have acted as glacial refugia for this species (3). Despite these findings, the southernmost distributional range of the species is not yet fully explored. With the aim of evaluating other potential diversity hotspots in this study we expand previous investigation, extending the sampling to other southernmost natural *T. aestivum* areas, such as Moroccan mountainous regions (Middle Atlas), southern Italy, and several Spanish and Balkan regions. The ongoing analyses based on haplotypes alignment of the ITS region of the rDNA, reveal as preliminary result a pronounced genetic variability among Moroccan *T. aestivum* populations, suggesting a possible phylogeographic differentiation also in these new areas of investigation. Additional analyses are underway to better trace natural populations of *T. aestivum* according to their geographic origin. The possibility of gaining more insight into the extent and distributional patterns of *T. aestivum* genetic diversity over its geographical range could be critical both for truffle industry's interest in selecting well-adapted inocula for the production of mycorrhizal seedlings, and for the establishment of programs devoted to biodiversity conservation.

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Changes In Red Data List Of Latvian Fungi (Wood Inhabiting Aphyllophorales) Based On IUCN Criteria.

Diāna Meiere [1,2]

[1]Institute of Biology, University of Latvia, Ojāra Vācieša str. 4, Riga, Latvia, diana.meiere@lu.lv,
[2]Latvian National Museum of Natural History, Kr. Barona str 4., Riga, Latvia, diana.meiere@lndm.gov.lv

The previous Red Data List of fungi of Latvia was developed as a part of the Red Data Book in 1996 (1). It contained 38 fungi species. Their conservation state assessments were based solely on expert opinion. In Latvia national legislation, requirements of protection of species and habitats are defined by the Law of Protection of Species and Habitats (ratified by the Parliament on 16.03.2000.). It contained 62 fungi species. In 2021-2023, during the development of the new Red Data List, species conservation state assessments were carried out accordingly to the standardized and worldwide used IUCN criteria (2). The outcome of this project will be an important and lasting step for fungal conservation. Currently, 112 species has been nominated to be red-listed in Latvia.

In the process of evaluation 13 species of wood inhabiting fungi remained in the red list but 28 were added to it. From the newly added species five are evaluated as critically endangered (*Amylocystis lapponica, Bondarzewia mesenterica, Hericium cirrhatum, Pseudoinonotus dryadeus* and *Pycnoporellus albuluteus*), 10 – endangered (*Anomoporia bombycina, Boletopsis grisea, Flavidoporia pulvinascens, Inocutis rheades, Inonotus leporinus, Sarcodonita crocea, Skeletocutis brevispora, Skeletocutis stellae, Trametes gibboa, Trametes suaveolens*) and 13 species – vulnerable (including *Abortiporus biennis, Flavidoporia mellita, Flaviporus citrinellus, Gloiodon strigosus, Inocutis dryophila, Junghuhnia pseudozillingiana, Punctularia strigosozonata, Rhodonia placenta, Sidera lenis, Skeletocutis odora*).

Fungi conservation state assessments were carried out accordingly to the IUCN guidelines for the Red List categories and criteria (IUCN Standards and Petition Committee, 2019). The distribution data was based on the species records, available at the Nature Conservation Agency database OZOLS, public nature observation portal dabasdati.lv, the website for the fungi of Latvia fungi.lv, and data of Latvian National Natural history museum fungi collection (nmkk.lv).

The work was done in the framework of LIFE project "Threatened species in Latvia: improved knowledge, capacity, data and awareness" (LIFE FOR SPECIES, LIFE19 GIE/LV/000857) with financial support of European Commission and the State Regional Development Agency, Republic of Latvia.

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Characterization of the microbial communities associated to blueberry plants in a natural environments.

Miriana Bortolot[1], Sonia Mazzarino[1], Alan Pizzinat[2], Bruno Sacchi[2], Silvia Perotto[1], Elena Martino[1,3] and Alessandra Salvioli di Fossalunga[1]

[1] Department of Life Sciences and Systems Biology, University of Turin, Italy;
[2] Joinfruit società consortile a.r.l.organizzazione produttori ortofrutticoli, Verzuolo (CN) Italy;
[3] Centre INRAE Grand-Est Nancy - Université de Lorraine - UMR 1136, 54280 Champenoux France

Plants are co-inhabited by a wide range of microorganisms such as bacteria, viruses and fungi that could be classified also depending on the plant compartment that they colonize. Their presence deeply affects plant's growth, health and stress resistance providing several vital services. Microorganisms application in agriculture as bioformulated products is a new emerging approach that could lead to a more sustainable agricultural system; however, to this aim, in-depth studies of the rhizosphere crop 'microbiota' are required [1-2]. Blueberry plants include different wild and cultivated species and during the last years they gained great commercial interest due to the numerous beneficial properties of their fruits on human nutrition and health [3]. These plants grow on acidic soils with good aeration and drainage, in habitats where essential nutrients are mostly present under organic form. The colonization of these harsh environments is mostly attributed to the capacity of these plants to establish an endosymbiosis with ericoid mycorrhizal (ERM) fungi. ERM fungi are versatile microorganisms, behaving both as saprotrophs and symbionts, and producing several extracellular enzymes which explain their ability to grow on complex substrates [4-5]. Despite the long commercial use and interest of these plants, few information are available about the composition and role of the blueberry associated microbiota. In this framework, our research aims were to provide an integrative view of the microbial communities associated with the blueberry plants and to analyze the potential role of their symbionts as plant growth-promoting microbes (PGPM). To address these goals, we characterized and compared the plant-associated fungal communities of *Vaccinium corymbosum* plants from two sampling sites, with different soils properties. Three different compartments, bulk soil, rhizosphere and endosphere at three different time points were considered performing a metabarcoding analysis. A culture-dependent approach to isolate members of the fungal community inhabiting internal root tissues was also performed, followed by the molecular identification of the selected isolates. From these analyses two new strains, corresponding to the archetypal ericoid mycorrhizal species *Hyaloscypha hepaticicola*, were obtained and tested in field trials with *V. corymbosum* plants, to investigate possible plant growth promotion abilities. The results obtained with these complementary approaches will be presented and discussed.

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Diversity of macrofungi in Samaria National Park and the Protected Areas of Western Crete – New records for the mycobiota of Greece.

Polemis, E.[1], Christodoulou, S.[1], Koutrotsios, G.[1], Paidi, C.[1], Vogiatzis, P.[1], Kontakos, D.[2] & Zervakis, G.I.[1*]

[1]Agricultural University of Athens, Laboratory of General and Agricultural Microbiology, Iera Odos 75, 11855 Athens, Greece; zervakis@aua.gr; [2]Management Unit of Samaria National Park and the Protected Areas of Western Crete, Old National Road Chanion-Kissamou, Agioi Apostoloi, P.O. Box 73100, Chania, Greece; mdpp.samaria@necca.gov.gr

The island of Crete is listed among the world's biodiversity hot-spots because of its long geological isolation and the existence of a mosaic of land habitats ranging from typical Mediterranean xerothermic to alpine, which explains the well-documented high endemism of plants and animals. Nevertheless, the fungal component of Cretan biodiversity still remains largely unexplored. Our ongoing research focuses on the inventory of mushroom species present in the Samaria National Park and in other Natura 2000 sites of western Crete. In this frame, more than 400 specimens were collected from various localities characterized by the dominance of *Pinus brutia* Ten, *Cupressus sempervirens* L., *Quercus ilex* L., *Q. coccifera* L. as well as by olive groves and abandoned fields. The material collected was subjected to detailed microscopical study, and the identification to species was achieved by combining morphological and molecular approaches. Among the findings, six species of Basidiomycota constitute new records for the Greek mycobiota. The recently described ectomycorrhizal *Boletopsis mediterraneensis* G. Moreno et al. and *Chroogomphus mediterraneus* (Finschow) Vila et al. were found growing in association with *P. brutia*. The xylotrophic *Neolentiporus squamosellus* (Bernicchia & Ryvarden) Bernicchia & Ryvarden, globally red-listed as 'endangered', was found on dead trunks of old-growth *C. sempervirens* trees. *Peniophora malenconii* Boidin & Lanq., another taxon with a strictly Mediterranean distribution, was collected from a dead trunk of the emblematic endemic tree species of Crete, i.e., *Zelcova ambelicea* (Lam.) Boiss. In addition, *Skeletocutis nemoralis* A. Korhonen & Miettinen, known so far from N. Europe, Iran and Japan, was found on a *Q. coccifera* branch. Last, the nitrophilous saprotrophic *Tricholomella constricta* (Fr.) Zerova ex Kalamees was collected from heavily grazed olive groves. The preliminary results presented here are indicative of the high diversity of macrofungi in Crete, and are suggestive of the need for further pertinent research in the island.

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Diversity of macromycetes in the island of Kefalonia (Ionian islands) – rare and interesting species.

Marina Triantafyllou[1], Zacharoula Gonou-Zagou[1]

Section of Ecology and Systematics, Department of Biology, National & Kapodistrian University of Athens, Panepistimiopolis, GR-15784 Athens, Greece, marintriant@gmail.com; zgonou@biol.uoa.gr

Kefalonia island, located west of mainland Greece (Ionian Sea) is notable for its many distinctive ecosystems and is dominated by the Mediterranean mountainous forest ecosystem of Mt. Ainos, which is the locus classicus of *Abies cephalonica*, the endemic fir of Greece. Aim of this work is the investigation of the diversity of macromycetes (mainly subphylum Agaricomycotina) of Mt. Ainos National Park and other localities in the island, as the existing data before this study were very few and fragmentary. Material was collected in Mediterranean mountainous fir forests to Mediterranean sclerophyllous forests during 16 field trips, at 16 different sites, from October 2012 to November 2020. More than 700 specimens were collected. The results of this study show that the diversity of macromycetes on the island is rich and comprises both common and rare species, as well as interesting specimens with intriguing taxonomic problems. *Mycopan scabripes* and thus the currently monotypic genus *Mycopan* are recorded for the first time from Greece. Additionally, eleven more taxa constitute new records from Greece, namely *Conocybe dumetorum*, *Geastrum corollinum*, *Lactarius mairei* var. *ilicis*, *Mycena pilosella* var. *heterocystidiosa*, *Mycena scirpicola*, *Mycena supina*, *Omphalina galericolor* var. *lilacinicolor*, *Phloeomana clavata*, *Porotheleum omphaliiforme*, *Pseudoclitocybe expallens* and *Resupinatus europaeus*. Moreover, *Clitocybula familia*, *Inocybe griseolilacina*, *Parasola lilatincta* and *Tulosesus subimpatiens* were recorded for the second time from Greece. For many species, such as *Atheniella adonis*, *Cantharellus alborufescens*, *Chamaemyces fracidus*, *Coprinopsis insignis*, *Cystolepiota seminuda*, *Exidia thuretiana*, *Lactarius mediterraneensis*, *Lepiota boudieri*, *Lepiota subgracilis*, *Lepiota subincarnata*, *Leucoagaricus crystallifer*, *Limacella delicata*, *Neolentinus cyathiformis*, *Parasola auricoma*, *Parasola kuehneri*, *Pluteus primus*, *Tricholomopsis flammula* and *Tubaria dispersa* very few nationwide reports exists. In parallel, many of the species that were identified during this study are currently being assessed under the ongoing project of the National Fungal Red List of Greece. The systematic study is still in progress and molecular methods are being used to unravel interesting taxonomic issues, concerning specific genera.

Dothideomycetes of Northern Ukraine Forests.

Tetiana V. Andrianova [1]

[1]M.G. Kholodny Institute of Botany, National Academy of Sciences of Ukraine, Tereshchenkivs'ka st., 2, Kyiv, Ukraine, andrianova.tetiana@gmail.com

Northern forests of the Central and Western Ukraine are the biodiversity centres. A few data on Dothideomycetes (Ascomycota) of the region were presented in the issued volumes of "Fungal Flora of Ukraine" (1980), database "Fungi of Ukraine" (1996) grounded on early investigations of Z. Girzhits'ka, T. Merezhko, M. Sokolova, V. Solomakhina, M. Tselle and M. Zerova. During the state project on fungi study in the western forests (2018-2022) a revision of KW mycological samples and new collections of Dothideomycetes as pathogens and saprotrophs, especially inhabiting leaves and herbs, was carried out. Summarized data represent the list of 543 species currently approved in 126 fungal genera. Fungi of 17 genera numbered 66,3 % of all revealed Dothideomycetes: *Septoria* – 74 species, *Diplodia* – 39, *Phyllosticta* – 30, *Ramularia* – 28, *Phoma* – 24, *Camarosporium* – 20, *Ascochyta* – 19, *Coniothyrium* – 17, *Alternaria* – 16, *Microdiplodia* – 14, *Cladosporium* – 13, *Leptosphaeria* – 13, *Mycosphaerella* – 13, *Hendersonia* – 12, *Cercospora* – 10, *Cucurbitaria* – 10, *Sphaerulina* – 8. Studied fungi are classified in Botryosphaeriales, Capnodiales, Catinellales, Cladosporiales, Dothideales, Hysteriales, Kirschsteiniotheliales, Mycosphaerellales, Myriangiales, Patellariales, Pleosporales and Venturiales; abundant were pathogenic fungi of the orders Botryosphaeriales (102 species of 10 genera), Mycosphaerellales (167 species of 27 genera) and Pleosporales (223 species of 63 genera). Fungi of Mycosphaerellaceae are represented by: *Caryophylloseptoria* (1 species), *Cercospora* (10), *Cercosporella* (1), *Exosporium* (1), *Fusoidiella* (2), *Mycosphaerella* (13), *Neocercospora* (1), *Neoovularia* (1), *Neophloeospora* (1), *Nothoseptoria* (1), *Paracercosporidium* (1), *Passalora* (4), *Phloeospora* (1), *Polythrincium* (1), *Pruniphilomyces* (1), *Pseudocercospora* (3), *Pseudocercosporella* (2), *Ragnhildiana* (1), *Ramularia* (28), *Rhabdospora* (5), *Rosisphaerella* (1), *Septoria* (74), *Sphaerella* (5), *Sphaerulina* (8), *Zasmidium* (1) and *Zymoseptoria* (1). New emerging epiphytotics of ordinary hemibiotrophs are registered for *Botryosphaeria visci*, *Cercospora armoraciae*, *Pseudocercospora opuli*, *Ramularia urticae*, *Septoria chelidonii*, *Sphaerulina gei* and *Wilsonomyces carpophilus* in natural habitats, according to the increase in average annual temperatures and droughts in the region. Good competitors in various forest biotopes were common species *Cladosporium herbarum*, *Coleroa robertiani*, *Diplodia mutila*, *Leptosphaeria ogilviensis*, *Microdiplodia coryli*, *Microsphaeropsis olivascens*, *Phyllosticta cruenta*, *R. coccinea*, *R. grevilleana*, *S. aegopodii*, *S. polygonorum*, *S. scabiosicola*, *S. trientalis* and *Sphaerulina westendorpii*. About 16% of Dothideomycetes species are rare and have been observed only once for Ukraine in this region. Investigations in the Rivne natural reserve, Prypiat'-Stokhid and Shats'k national nature parks revealed unknown for the Ukrainian mycobiota, mainly Eurasian species of *R. libanotidis*, *S. balloticola*, *S. dysentericae* and *S. hydrocotyles*, and other microfungi as *Asteroma solidaginis*, *Colletotrichum acutatum*, *Neoramularia bidentis*, *Phomopsis vaccinii*, *Pestalozziella subsessilis* (Andrianova, 2020; Андріанова, 2020, 2021 a,b; Андріанова, Ковальчук, 2021). Noted first records of the rare fungi known from the south and east of Ukraine: *Cl. galii*, *R. crepidis*, *R. cupulariae*, *R. gnaphalii*, *R. heraclei*, *R. lysimachiae*, *R. simplex*, *Rhabdospora visci*, *S. callae*, *S. lycopi*, *S. tabacina*, in the western forests.

Involvement of miRNA and siRNA in multidirectional communications in wheat-pathogenic *Fusarium culmorum* and wheat-beneficial *Trichoderma atroviride* interactions – a preliminary study.

Sylwia Salamon, Polina Havrysh[1], Lidia Błaszczyk[1]

[1]Department of Plant Microbiomics, Institute of Plant Genetics Polish Academy of Sciences, Poland;
ssal@igr.poznan.pl; phav@igr.poznan.pl; lbla@igr.poznan.pl

The interactions between plants and their associated fungi are complex and the consequences diverse. Fungi living in close association with plants are known to be beneficial, neutral, or pathogenic. Nevertheless, the molecular mechanisms of plant communication with their interacting fungi are still poorly understood. In order to gain insights into these interactions, there is first the need to understand the complex interplay of signals between fungi and host plants, to decode the functions of both microbial and plant signals and their respective receptors, as well as their roles in triggering defensive and immune responses in plant. The essential components of this complex network of gene regulatory pathways have been found to be the endogenous small RNAs (sRNAs): micro RNA (miRNA) and small interfering RNA (siRNA). These molecules act as specific repressors of target gene expression, either at the transcriptional level by DNA methylation and/or histone modification, or at the post-transcriptional level by cleavage of the target transcript or inhibition of translation. Wheat (*Triticum aestivum* L.) plays an important role in maintaining global food security. In order to assess the possibility of miRNA involvement in the interactions of wheat with pathogenic *Fusarium culmorum* and beneficial *Trichoderma atroviride* fungi, three miRNAs (miR398, miR167 and miR159) were selected. The absolute quantification of these mature miRNAs as well as their potential target transcripts were determined using the digital PCR approach. It was observed that the expression profiles differ spatially (aerial and underground organs), temporally (early and late reactions), and also depend on the species of the fungus. Moreover, we noticed that the response of plants at the miRNA level during fungal infection was higher in the early phase of *F. culmorum* infection and dynamically decreased several days after inoculation, which may suggest a role of studied miRNAs in neutralization of hormonal imbalance and oxidative stress caused by fungal invasion, and consequently initiating a host defense response via the signaling pathway. Furthermore, to assess the involvement of DNA methylation in genes participating in wheat-fungi interactions, we determined the methylation profiles of promoter regions of *TIFY9* (coding Jasmonate ZIMdomain protein) and *ERF6* (Ethylene-responsive transcription factor6) genes. We conducted DNA pyrosequencing after bisulfite conversion in leaves of two wheat cultivars (resistant and susceptible to fusariosis), 48 hours after inoculation with the aforementioned fungi. We observed that expression of *TIFY9* is controlled by DNA methylation mechanism. *TIFY9* promoter region is methylated in all studied groups, however the amount of methylated cytosines are significantly increased after *T. atroviride* inoculation compared to *F. culmorum* ($p=0.05$) and control ($p=0.07$) groups. Contrary, promoter region of *ERF6* gene is unmethylated regardless of wheat cultivar and applied conditions**.** The results of preliminary tests indicate the importance of epigenetic mechanisms in control of transcriptional reprograming under the beneficial and harmful wheat-fungi interactions and demonstrate the needs to improve our current understanding of this regulations.

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Endophytic fungi isolated from wheat tissues influence changes in its transcriptome.

Polina Havrysh[1], Sylwia Salamon[1], Lidia Błaszczyk[1]

[1]Institute of Plant Genetics, Polish Academy of Sciences, 34 Strzeszynska Street, 60-479 Poznan, Poland, phav@igr.poznan.pl, ssal@igr.poznan.pl, lbla@igr.poznan.pl

Due to the changing climate and related weather anomalies, wheat crops are exposed to an increased number of combinations of abiotic and biotic stresses that pose a threat to the production of this economically important grain. Agricultural producers can observe the negative effects of climate change, manifested by reduced water availability during the growing season, or more frequent and intense heat waves. In addition, climate change causes the emergence of new pathogens, including pathogenic fungi, which, by causing diseases, significantly reduce the quantity and quality of grain.

To be able to propose new, safe, and ecological approaches to protect wheat against emerging threats, based on biological methods, it is important to understand the mycobiome (mycobiota associated with) of wheat plants, especially the endosphere, and the interaction of endophytic fungi with the host. One of the levels of these interactions may be indicated by the response of plants to endophytic fungi at the transcriptome level.

Therefore, the aim of the present study was to analyze the transcriptome of common wheat seedlings (Polish wheat cultivar Euforia) after inoculation with fungi previously isolated from the wheat endosphere, using the RNA-seq technique (HiSeqX, Illumina) and verified by RT-qPCR. In this work, transcripts with a statistically significant difference in expression were selected. It was shown that the least effect on wheat seedlings was observed after inoculation of plants *Sarocladium strictum* strains: only 4 transcripts were overrepresented and 6 showed reduced levels compared to the control. Inoculation of *Sarocladium spinificis* and *Chrysosporum pseudomerdarium* resulted in similar numbers of overexpressed transcripts (475 and 588, respectively), with treatment of *Ch. pseudomerdarium* resulted in a down-regulation of 604 transcripts and *S. spinificis* – down-regulation of 125 transcripts. It was shown that differentiating transcripts after inoculation of *S. spinificis* are mainly involved in the metabolic (24.5%), cellular (28.3%) and stimuli (20%) responses, and after inoculation of *Ch. pseudomerdarium* in cellular (22.5%), metabolic (12.5%), developmental (10.8%), biological regulation (9%), localization (8%), regulation of biological processes (7%). The greatest impact on the changes in the wheat transcriptome had the treatment of wheat by *Penicillium olsoni* endophytic strain - as many as 6939 transcripts showed a reduced level of expression compared to the control, while 4124 transcripts were overexpressed. It was noted that significantly differentiating transcripts participate in the following biological processes: cellular (30.5%), metabolic (21.5%), biological regulation (9.59%), response to stimuli (9.3%), biological (7.56%). Selected significantly differentially expressed transcripts were verified using the RT-qPCR method. Potentially, these transcripts can be considered in terms of their participation in the reaction of wheat to strains of endophytic fungi. Molecular mechanisms can provide information on the role and potential function of endophytic species, and in the longer term, their participation in the modulation of the microbiome of wheat plants in the context of crop health protection.

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Endophytic fungi of *Humulus lupulus* as a potential source of biocontrol agents.

Claudia Riccioni[1], Andrea Rubini[1], Maurizio Cenci[1], Cristina Vettori[2], Donatella Paffetti[3], Beatrice Belfiori[1]

[1]National Research Council (CNR), Institute of Biosciences and Bioresources (IBBR), Division of Perugia, Via della Madonna Alta, 130, 06128 - Perugia (Italy) E-mail: claudia.riccioni@ibbr.cnr.it, andrea.rubini@ibbr.cnr.it, maurizio.cenci@ibbr.cnr.it, beatrice.belfiori@ibbr.cnr.it
[2]National Research Council (CNR), Institute of Biosciences and Bioresources (IBBR), Division of Florence, Via Madonna del Piano, 10, 50019 – Sesto Fiorentino (FI) Italy. E-mail: cristina.vettori@ibbr.cnr.it [3]Dipartimento di Scienze e Tecnologie Agrarie, Alimentari, Ambientali e Forestali (DAGRI), Università degli Studi di Firenze, Piazzale delle Cascine, 18 – 50144, Italy. E-mail: Donatella.paffetti@unifi.it

Hop (*Humulus lupulus* L.) is a dioecious plant of the *Cannabaceae* family that grows spontaneously in temperate zones and is cultivated for the brewing industry, as well. In fact, its secondary metabolites, occurring in the female flowers (cones), provide flavor, bitterness, aroma and antimicrobial properties to the beer. In addition, some metabolites are bioactive substances with sedative, antimicrobial and anticancer properties (1,2). Wild hop germplasm is needed to provide new genetic resources for hop breeding, since, to remain competitive in the hop industry, hop breeders must respond to the ever-changing needs of the brewing community by providing appropriate new cultivars (3) with novel aromatic bouquets and/or better agronomic performances. Therefore, in order to select genetic materials useful to improve the commercial hop germplasm, European wild hops have been screened in the last decade, including Italian accessions, that have been analyzed by genotyping and phytochemical profiling (4). Although Italy is not known for growing hops, in recent years, there is a growing interest in hop cultivation that started in small farms and in experimental fields, due to the rising number of microbreweries that aim at using locally grown rather than imported hops. However, several diseases and pests can really compromise the hop production. The scarcity of plant protection products registered for hops in Italy severely limits the possibility to control diseases, thus hop protection management must focus on proper agronomic practices and biostimulants for induction of plant resistance (5). Plant endophytic fungi represent a valuable resource of potential biocontrol agents for the management of plant diseases; they help in reducing the environmental impact of crop plants as they allow the reduction in agrochemicals and fertilizers. The aim of this work was to characterize and estimate the diversity of the fungal endophytes associated with different tissues (roots, stems, leaves, cones and male flowers) of *H. lupulus* wild accessions collected in different sites of central Italy. We adopted a double approach: a classical isolation-based approach allowed us to build a strain collection available for the screening and identification of biocontrol agents against plant pathogens; in parallel a metabarcoding approach based on the massive sequencing of the ITS2 region using the Ion-Torrent platform is ongoing to get a more comprehensive picture of the endophytic fungi communities inhabiting the different tissues, including both colturable and not-colturable strains. Experiments to identify a first set of potentially bioactive fungal strains against phytopathogenic fungi and comparisons among the community structure of the different tissues in the different collection sites are under progress.

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Epiphytic fungal antagonists as biocontrol agents of *Alternaria* leaf spot of *Brassicas*.

Jovana Blagojević[1]

[1] Institute for Plant Protection and Environment, Teodora Drajzera 9, Belgrade, Serbia, jovanaizbis@gmail.com

Plants of the genus *Brassica* occupy the top place among vegetables in the world with a steady trend of increasing yields and growing area (1). Brassica crop losses due to leaf spot disease caused by Alternaria pathogens, specifically *A. brassicicola*, are reported worldwide and are often controlled by chemical fungicides (2). During the five-year study of Alternaria spp. in various *Brassica* host plants in Serbia, highly virulent strains of *A. brassicicola* were detected. In the same study, fungi were isolated from the phyllosphere of healthy Brassica plants in Alternaria disease-affected fields and tested against disease. Two epiphytic fungal strains with high antagonistic activity were detected from Brassicas leaves and identified as *Trichoderma* sp. and *Fusarium* sp. The objective of this study was to investigate their antifungal potential to *A. brassicicola* isolates, *in vitro* and *in vivo*. For *in vitro* test, the dual culture method was used and the colony diameter of *A. brassicicola* isolates was measured after 7 days of growth on PDA at 25 °C, and the percent inhibition (I) relative to the control was calculated. Both antagonists revealed a high inhibition effect on the growth of *A. brassicicola* with *Trichoderma* sp. being more effective than *Fusarium* sp. An *in vivo* test was performed on healthy eight-week-old cabbage plants in two ways. First, leaves were treated with a spore suspension of *Trichoderma* sp. or *Fusarium* sp., and 24 h before spraying pathogen spore suspension. The second test involved applying a pathogen spore suspension on leaves 24h prior to the spraying of the antagonists' spore suspension. The antagonist efficacy, measured as a percentage of necrotic leaf surface relative to the control, was significantly higher when leaves were first treated with the antagonists' spore suspension and no significant difference in action between *Trichoderma* sp. and *Fusarium* sp was reported. Given that cabbage cultivars resistant to Alternaria pathogens have not been registered so far, biological control of *Alternaria* spp. represents an essential strategy of their suppression that should be integrated into management strategies.

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Exploring taxonomic and functional diversity of forest soil fungi in central Italy by metagenomic analyses.

Andrea Rubini[1], Beatrice Belfiori[1], Cristina Vettori[2], Donatella Paffetti[3], Claudia Riccioni[1]

[1]National Research Council (CNR), Institute of Biosciences and Bioresources (IBBR), Division of Perugia, Via della Madonna Alta, 130, 06128 - Perugia (Italy) E-mail: andrea.rubini@ibbr.cnr.it, beatrice.belfiori@ibbr.cnr.it, claudia.riccioni@ibbr.cnr.it
[2]National Research Council (CNR), Institute of Biosciences and Bioresources (IBBR), Division of Florence, Via Madonna del Piano, 10, 50019 – Sesto Fiorentino (FI) Italy. E-mail: cristina.vettori@ibbr.cnr.it
[3]Dipartimento di Scienze e Tecnologie Agrarie, Alimentari, Ambientali e Forestali (DAGRI), Università degli Studi di Firenze, Piazzale delle Cascine, 18, 50144, Italy. E-mail: Donatella.paffetti@unifi.it

Fungi have a fundamental role in biogeochemical cycles and are one of the major pools of living biomass in forest soil ecosystems. Besides their fundamental role in organic matter degradation and recycling, fungi represent an important source of ecosystem services. Fungal assemblages in forest environments are influenced by several factors such as climate, soil characteristics and interaction with host plants and other microorganisms such as bacteria and archaea. In turn, the fungi may influence plant health, stress response and can either act as pathogens or as a defense against pathogens. Furthermore, several fungi (especially macro-fungi) are harvested and/or cultivated as edible mushrooms and as species with medicinal or biotechnological applications (1). Moreover, composition of fungal macro- and micro-communities and their variation through the years may represent valid bioindicators to study the effect of climate changes on terrestrial environments (2). Fungal biodiversity studies are typically performed by long term surveys of fruit bodies production and their taxonomic characterization based on morphological and/or molecular techniques. More recently the development of next generation sequencing technologies has allowed performing studies based on DNA metabarcoding by analyzing target metabarcodes such as the ITS region of rDNA, or by shotgun metagenomic sequencing. Several studies revealed that the fungal species producing fruit bodies or those that are culturable in vitro represent only a small fraction of the total biodiversity. Thus sequence-based studies have been revealed much more informative in disclosing the true species diversity. Metagenomics has revealed also a great potential for bioprospecting new enzymes and bioactive molecules directly from sequencing, without the necessity of isolation (3). Here, we analyze the taxonomic and functional diversity of fungal and bacterial communities present in two representative forest biomes in central Italy: beech forests (*Fagus sylvatica*) and oak forests (*Quercus cerris*). Ion Torrent PGM technology was used to sequence metabarcoding libraries based on ITS and 16S markers for fungi and bacteria, respectively. Functional diversity was also assessed by performing shotgun metagenome sequencing of total soil DNA using the Oxford Nanopore Minion technology. The main aims of this work were to study the fungal taxonomic and functional diversity in forest ecosystems of central Italy and find correlations between fungal and bacterial assemblages. Metagenomic sequence data were also explored to retrieve fungal and bacterial sequences of potential biotechnological interest such as lectins, cellulases, lipases.

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First record of five species of genus *Tuber* in North Macedonia.

Slavica Tofilovska[1,2], Katerina Rusevska[1,2], Tome Jovanovski[2], Mitko Karadelev[1,2]

[1]Institute of Biology, Faculty of Natural Sciences and Mathematics, Ss Cyril and Methodius University, Arhimedova 5, Skopje, North Macedonia, stofilovska@gmail.com
[2]Macedonian Mycological Society, Arhimedova 5, Skopje, North Macedonia

Up until now, in North Macedonia 18 taxa within genus *Tuber* are published in the checklist of Ascomycota for the country, *T. aestivum*, *T. aestivum* var. *uncinatum*, *T. bellonae*, *T. borchii*, *T. brumale*, *T. brumale* var. *moschatum*, *T. dryophillum, T. excavatum, T. foetidum, T. fulgens*, *T. macrosporum, T. magnatum, T. melanosporum, T. mesentericum, T. puberulum, T. rufum* var. *rufum* f. *ferrugineum, T. rufum* var. *rufum* f. *lucidum*, *T. rufum* var. *rufum* f. *nitidum* (1). Investigations on the diversity of hypogeous species in North Macedonia was deepened since 2019, a review of deposited collections in the Macedonian Collection of Fungi (MCF) was performed along with retrieving of new materials. Unknown species for the country were found and following a detail morphological analysis, the presence of *T. anniae, T. maculatum, T. oligospermum, T. rapaeodorum* and *T. regianum* was confirmed. The distribution is shown by their presence in the defined biogeographical regions of the country (2). *T. anniae* is a rare species in Europe, confirmed only in Finland, in North Macedonia it was found in *Picea abies* forest at 1800 m.a.s.l. on the mountain Shar Planina. *T. maculatum* and *T. rapaeodorum* were discovered in riverine habitats. *T. maculatum* in association with *Populus alba* and *Salix alba* at 400 m.a.s.l. in the valley Polog, while *T. rapaeodorum* with *P. alba* at 400 m.a.s.l. on mountain Karadzica. The mixed forest of *Quercus pubescens* and *Carpinus orientalis* are widely distributed in the country and well researched for truffles, however *T. oligospermum* is also a rarely encountered species which was found only on the mountain Jakupica at 600 m.a.s.l. *T. regianum* was noted only once in *Fagus sylvatica* forest, mountain Bistra on 1100 m.a.s.l. Maybe the small size of the sporocarps is reason for omission of this species in the frequent research in *F. sylvatica* forests. The diversity of true truffles in the country is under study with molecular analyses and up until know it is apparent that one of the more frequently represented phylogenetic groups is the puberulm clade.

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Five new fungi records from Shar Planina National Park in Macedonia.

Hasime Jashari[1], Slavica Tofilovska[1], Mitko Karadelev[1], Katerina Rusevska[1]

[1]Institute of Biology, Faculty of Natural Sciences and Mathematics, Ss. Cyril and Methodius University, Arhimedova 5, Skopje, Macedonia, jasharihasime@gmail.com

Shar Mountain is one of the largest mountain massifs in Macedonia, distinguished by outstanding species diversity, first and foremost owing to the presence of various climate and vegetation features. Recently, it was proclaimed as a national park. Published data on fungi of Shar Mt is scarce, they refer to specific areas, and there is not any systematic data available on fungi occurrence in different forest associations. Therefore, we decided to embark on mycodiversity study of the coniferous communities in this mountain massif.

Research was implemented in the coniferous forests on the territory of Shar Mt, at an altitude ranging between 750 m and 1,805 m.

The material was collected from localities with natural coniferous wood, Popova Shapka, Jelak, Leshnica and the Adzhina Reka river, Shipkovica, and also from pine plantations from the vicinity of Rogachevo village. Fungi were collected from various substrates, such as soil, fallen branches, stumps, old trunks, trees, etc. Some specimens were identified in the field whereas others were determined in the Mycological Laboratory.

The study resulted in 184 fungi species recorded. However, when taken into account data from literature the total number of macromycetes from coniferous communities of Shar Mt is 207 species. The phylum Basidiomycota comprises 197 species (whereof 6 are gasteroid fungi); 7 species are affiliated to Ascomycota (2 of them are lichens), and only one species belongs to the phylum Myxomycota. The greatest part of fungi species was observed in the coniferous communities of Jelak (97). Forty-four species were noted in the fir-spruce association of the Adzhina Reka river whereas 8 species were recorded at Popova Shapka. Eight species were identified in the pine plantations in Rogachevo, and 4 species in the mixed fir-oak association (Shipkovica). Some species are used for human nutrition, such as the following: *Amanita rubescens, Boletus edulis, Calvatia utriformis, Russula cyanoxantha, Suillus granulatus, S. luteus.* The species *Hericium coralloides* is on the Macedonian Red List of Fungi, in the category of endangered species. It is worth noting the finds of the species *Agaricus altipes* (=*A.aestivalis*), *Cortinarius flexipes, C. triformis, Lyophyllum konradianum* and *Pluteus primus,* which are first records for the country.

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Fruiting pattern and habitats of protected ascomycete *Sarcosoma globosum* in Lithuania.

Eglė Vabuolė[1], Sigitas Juzėnas, Ernestas Kutorga

[1]Institute of Biosciences, Life Sciences Center, Vilnius University, 7 Saulėtekio av., Vilnius, Lithuania, egle.semaskaite@gmc.stud.vu.lt

The ascomycete *Sarcosoma globosum* is rare and declining species. The shrinkage of its geographical distribution range was observed in Central Europe. Known localities of this fungus in Lithuania are situated on the southwestern edge of its current distribution in Europe. Past, ongoing, and future habitat declines are estimated to impact negatively on all populations of *S. globosum* (1). The reproduction ability of this fungus in changing habitat and climate conditions is not fully known, therefore, the aim of our investigation was to assess the fruiting pattern and habitats of protected ascomycete *S. globosum* in Lithuania. The studies were caried out from 2007 (1 known locality) till 2023 (28 localities). In total, 8 654 fruit-bodies of *S. globosum* have been counted during vernal field surveys of study sites. Fungus exhibit significant yearly quantitative fluctuations. The annual fruit-body count in a single locality varied from 0 to 926. The annual mean number of fruit-bodies per locality varied from 1 (in 2007) to 114.2 (in 2015), the annual production average for 2007–2023 period was 18.2 fruit-body per locality. The fungus usually starts fruiting early in the spring, nevertheless, the earliest ever start of *S. globosum* fruiting season in Lithuania was registered on the 2nd of November in 2015. The development of individual fruit-bodies has been studied in 2021–2023. Duration of fruit-bodies varied from 2 to 7 months. Vast number of fruit-bodies do not reach the maturity (sporulation) – 54.6% of the fruit-bodies matured in 2022 and only 31.7% – in 2023. In general, the fungus *S. globosum* inhabits both planted monoculture Norway spruce plantations and naturally regenerated spruce or spruce-pine forests, has a preference to denser canopy and understory coverage, and prefer intact moss layer and forest litter. Majority of *S. globosum* growth sites (58.1%) are situated in forests over 71 years old, however, large proportion (25.6%) of sites are located in young spruce stands. The computed value of stand density index varied from 231.5 to 1573.9, and the stand basal area values varied from 11.4 m^2/ha to 468.8 m^2/ha. The pH value of organic soil layer varied from 3 to 6, and C:N ratio varied from 11.1 to 20.2. Disturbances (windthrow, snowbreak, tree damage by pests, forest management activities, forest floor disturbance) were studied in fungus habitats. Only 11.6% of *S. globosum* localities are situated in strictly protected territories.

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Fungi associated with *Tomicus minor* infesting Scots pine forests in Slovakia.

Michaela Strmisková[1,2], Marek Barta[1], Miriam Kádasi Horáková[1], Katarína Pastirčáková[1]

[1]Slovak Academy of Sciences, Institute of Forest Ecology, Department of Plant Pathology and Mycology, Akademická 2, SK-94901 Nitra, Slovakia, strmiskova@ife.sk, marek.barta@savba.sk, kadasi@ife.sk, katarina.pastircakova@ife.sk; [2]Technical University in Zvolen, Faculty of Ecology and Environmental Sciences, T.G. Masaryka 24, SK-96001, Zvolen, Slovakia, xstrmiskova@is.tuzvo.sk

In Slovakia, Scots pine (*Pinus sylvestris*) is the second most important coniferous species representing about 7% of the total afforested area in the country. Over the last decade, there has been a persistent decline in the health of pine stands across almost the entire territory of Slovakia. Climatic extremes and the annual increase in the population density of several scolytid species create favourable conditions for the spread of fungal pathogens. Bark beetle outbreaks occur mainly in the Záhorie region, where the current situation is calamitous and very unfavorable for the stability of pine stands. Most bark beetle species are closely associated mainly with ophiostomatoid fungi, which cause blue staining of wood, as well as serious diseases of host trees. In Slovakia, detailed studies on ophiostomatoid fungi that parasitize pine trees have not yet been undertaken. In the spring of 2022, a survey was carried out to determine fungi that are associated with the lesser pine shoot beetle (*Tomicus minor*) in Scots pine forest stands located in western Slovakia. Individual beetles were collected from separate galleries using sterilized forceps and placed individually into sterile microtubes. Fungi from the beetles were isolated according to standard isolation procedures on 2% malt extract agar supplemented with cycloheximide, streptomycin sulphate and tetracycline hydrochloride, followed by cultivation under controlled conditions in a growth chamber. The isolated pure cultures were grouped based on their morphology. ITS, beta-tubulin and elongation factor 1-alpha sequences were determined for isolates representing each morphological group. Ten *Ophiostoma* species (*O. breviusculum*, *O. canum*, *O. floccosum*, *O. fuscum*, *O. ips*, *O. macroclavatum*, *O. minus*, *O. nitidum*, *O. piceae*, *Ophiostoma* sp.) and *Leptographium piceaperdum* were isolated from the beetles, along with common saprotrophic fungi (*Alternaria alternata*, three *Cladosporium* species, *Mucor* sp., six *Penicillium* species, *Stemphylium* sp.) and entomopathogenic fungi (*Beauveria bassiana*, *B. pseudobassiana*, *Clonostachys rosea*, *Lecanicillium fusisporum*, *L. psalliotae*). *Ophiostoma minus* and *O. canum* were the most prevalent ophiostomatoid species. The results of this study have contributed to a better understanding of the fungi associated with pine-infesting bark beetles in Slovakia.

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Fungi of Antarctica.

Veronica Karmanova[1]

[1]V.F. Kuprevich Institute of Experimental Botany of the National Academy of Sciences of Belarus, 27, Akademicheskaya str., 220072, Minsk, Republic of Belarus, veronikarmanova@gmail.com

Antarctica is the coldest, driest and highest continent on our planet. Antarctic ecosystems are unique, because they are characterized, firstly, by a wide range of environmental factors that are extreme for living organisms, and, secondly, by a relatively minimal anthropogenic load. Fungi are the most diverse group of microorganisms that colonize Antarctic substrates of natural and anthropogenic origin. Fungi in Antarctica are found in two forms - filamentous fungi and yeasts. They have a high degree of ecological plasticity, which allows them to survive in extreme conditions such as extreme temperature changes (including extreme low temperatures), high levels of UV radiation, constant freeze-thaw cycles, dehydration, osmotic stress and low nutrient concentrations. Fungal communities in Antarctica play a role in nutrient and carbon cycling in terrestrial and marine ecosystems, participate in soil formation and in situ transformation of rocks, and act as symbionts, mutualists, pathogens and saprotrophs that form a complex biological network with other organisms, and are an interesting model for studying the coevolution of symbiosis under extreme conditions (1). The study of the mycobiota of Antarctica began in the 1960s. To date, representatives of the phyla Ascomycota, Basidiomycota, Mucoromycota, Chytridiomycota, and Glomeromycota have been identified in Antarctica, as well as mushroom-like Oomycota and Mycetozoa. A distinctive feature of the fungi of Antarctica is the predominance of the Ascomycota phylum, and not Basidiomycota, as on other continents. Initially, cultural and morphological methods were used to study fungal diversity. Later, molecular biological methods (analysis of ITS regions, sequences of mitochondrial genes, and protein-coding nuclear genes) became widely used. Today, metagenomic analysis is used to study the taxonomic spectrum of micromycetes. The most common species in Antarctica are the genera *Thelebolus*, *Cylindrocarpon*, *Glomerella*, *Golovinomyces*, *Penicillium*, *Phoma Cadophora*, *Geomyces*, *Rhodotorula; Cladosporium*, *Cladophialophora; Antarctomyces*, *Hyphozyma*, *Goffeauzyma*, *Geotrichum*. Strong selective pressure leads to the development of new mechanisms of stress resistance, implemented at the physiological and biochemical levels. This is expressed in the ability to produce a variety of specific compounds (antibiotics, enzymes, pigments, toxins) that provide adaptation and increased competitiveness. In addition, biochemical studies carried out on fungi isolated from various Antarctic environments have shown that, compared to mesophilic fungi, they synthesize metabolites and enzymes with atypical structures and are potentially one of the most promising producers of new compounds (2). To sum up, the study of the biological diversity of micromycetes of Antarctica is important both in the fundamental aspect, for establishing the role of fungi in extreme ecosystems and identifying the patterns of their distribution in the biosphere, as well as for studying the adaptation mechanisms of eukaryotic organisms to extreme environmental factors, and in the applied aspect, to identify potential producers of biologically active substances.

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Fungi of Kamchatka peninsula volcanic soils: metabolism and diversity. Micromycetes inhabiting volcanic soils: metabolism and diversity.

Anna Shestakova[1], Daria Surkova[1], Elizaveta Ganetskaya[1], Anjelika Filippova[1], Irina Ognenovska[1], Osmolovskiy Alexander[1]

[1]HSE University, 101000 Moscow, Russia, aashestakova_4@edu.hse.ru

Kamchatka is a peninsula on the northeast of Eurasia, and 40% its landscape is covered with volcanoes. They have been forming poor black soils rich in metal oxides with low organic content. Micromycetes of Kamchatka and their metabolism are poorly studied. Fungi are destructors of organic matter and studying the inhabitants of poor soils is interesting and may lead to finding new oligotrophic or highly-tolerant species. 13 strains of microscopic fungi were isolated from samples of Kamchatka volcanic soils collected in August, 2022. They were represented by conventional *Alternaria*, *Aspergillus*, *Penicillium*, *Fusarium* and *Mortierella* species, as well as less frequently occurring *Purpureocillium*, *Metharisium*, and *Pseudogymnoascus*. For the isolated species, enzyme production, growth rates, and antibiotic activity was assessed. Using agarized media with corresponding substrates, it was shown that among studied, most species were capable of producing amylases, lipases, cellulases, and pectinases. These substrates are easily found in soils, as plants can colonize even poor habitats. Half of the isolates were capable of protease production. *Pen. brevicompactum*, *Pen. aurantiogriseum*, *F. pallidoroseum*, and *Ps. pannorum* were found to be active proteolytic, preferring fibrous collagen rather than globular casein. This fact is needed to be explained, since usually fibrous proteins' degrading enzymes are less frequently observed for microorganisms. For *Ps. pannorum*, no proteases were previously described, so this species should be further investigated as a protease producer, e.g., for potential industrial application. Four isolates (of genera *Aspergillus, Penicillium, Metharisium* and *Fusarium*) expressed chitinolytic activity, which indicated that these fungi can invade insects or mollusks, or decompose their residues. Such known enthomopathogenic species as *Met. anisoplea* and *Pur. lilacinum* might have been associated with actively moving animals (e.g., flying mosquitoes), brought to the area with volcanic soils, and remained there. Most of the strains also produced organic acids, hereby lowering the pH value of their local environment. This may act as an antagonism factor, as fungi are generally acidophilic, and many bacteria are not, as well as an adaptation to the high concentrations of bounded metals in soil. Only one strain (*F. pallidoroseum*) produced alkaline metabolites, thus, increasing pH. Among isolated, three species exhibited antimicrobial activity: *A. niger*, *Pen. solitum* (against Gram-positive and Gram-negative bacteria), and *Mor. Polycephala* (against Gram-positive bacteria). When *Aspergillus* and *Penicillium* fungi are known as antibiotic producers, for *Mor. polycephala* no antibiotics were described. Thus, mycobiota of volcanic soils has a hidden potential for both fundamental and applied research.

Interesting and unusual corticioid Fungi (Basidiomycota) from the Iberian Peninsula.

Isabel Salcedo[1], Ibai Olariaga[2], Ireneia Melo[3]

[1]Department of Plant Biology and Ecology, University of Basque Country, (UPV/EHU), Sarriena s/n, 48940 Leioa, Bizkaia, Spain, isabel.salcedo@ehu.eus; [2]Department of Biology and Geology, Physics and Inorganic Chemistry, University Rey Juan Carlos, Móstoles, Madrid, Spain, ibai.olariaga@urjc.es; [3]University of Lisboa, National Museum of Natural History and Science / Centre for Ecology, Evolution and Environmental Changes, Natural History and Sistematics (NHS) Research Group (cE3c), Lisboa, Portugal, mimelo@ciencias.ulisboa.pt

Distribution of corticioid fungi are influenced by conditions for fungal establishment and growth related to climate and host tree distribution, as well as to dispersal dynamics at various scales in space and time. The Iberian Peninsula has a great richness and diversity of woody substrates for the development of these fungi. Large areas of beech, oak and even pine forests, as well as their replacement heaths, are found in the Atlantic region, while in the Mediterranean region sclerophyllous, semi-deciduous forests and shrublands dominate, with different pine species in the mountainous areas. Due to this great diversity of substrates and its geographical location, some interesting species of corticioid fungi extend their distribution to the Iberian Peninsula. *Aleurodiscus wakefieldiae* Boidin & Keller, a species growing on dead hardwood, with rare occurrences in Europe, in the south of England and southwest of France, extends its distribution area to the north of Spain, to the arc of the Bay of Biscay, in the Basque Country. It was also found in Asia, in Taiwan, and in North America (Pennsylvania, USA). *Coniophora ladoi* Tellería, a controversial species found on dead coniferous wood, only reffered to Spain, in the type locality (Galicia), and more recently collected in Somiedo (Asturias). *Cunninghammyces umbonatus* (G. Cunn.) Stalpers, a species described from New Zealand, restricted to species of Apiaceae, and also found in eastern Australia, Asia (Taipei), Reunion Island, South America (Venezuela) and Europe (France, Switzerland, Portugal and Spain), always rare. *Dendrothele griseocana* (Bres.) Bourdot & Galzin, always found on bark of living deciduous trees, is a species widely distributed but little known in Europe, reported from the Mediterranean (including Portugal) and north to Denmark. *Dentipratulum bialoviesense* Domański, described to grow on decayed wood of *Abies*, and only registered in Poland and France, was now found in the Basque Country (Spain), growing near or on old polipore species growing in pine. *Penttilamyces olivascens* (Berk. & M.A. Curtis) Zmitr., Kalinovskaya & Myasnikov, a species widely distributed but very rare in Europe, found on decayed wood. It is known from France, Belarus and now reported from Portugal and Spain. Outside Europe it was reffered to India, Japan, North America and West Indies. *Sistotrema autumnale* Ryvarden & H. Solheim, species with a worldwide distribution, reported from northern and central Europe, was now collected in Spain. *Trechispora kavinioides* B. de Vries, species growing on decayed coniferous wood, widely distributed in Europe but unknown in the Iberian Peninsula, was recently collected in the North of Portugal.

Lichens as a habitat for microfungal communities.

Marina Temina[1], Isabella Grishkan[2]

[1]Institute of Evolution, University of Haifa, 199 Aba Khoushy Ave, Mount Carmel, Haifa, IL-3498838, Israel, E-mail marina@evo.haifa.ac.il; [2]Institute of Evolution, University of Haifa, 199 Aba Khoushy Ave, Mount Carmel, Haifa, IL-3498838, Israel, E-mail bella@evo.haifa.ac.il

Lichen thalli as a substrate harboring specific endolichenic fungal communities, has attracted the attention of mycologists during the past decades. Studies dealing with endolichenic fungi have been performed in different geographical regions and ecosystems. These studies included various lichen species on a variety of substrates, and implied both culture-based and culture-independent molecular methodologies. Nevertheless, lichen thalli are still considered mycologically poorly explored habitats where novel fungal taxa can be found. In Israel, endolichenic fungi in the thalli of saxicolous lichens have been studied in three different regions. Two sites were chosen in the Mediterranean part of Israel, one of them was in the Alma–Har-Ben-Zimra area (AZ) in the Upper Galilee, and the second site was in the Nahal Oren valley (NO) in the Mount Carmel. The third site was located in the central Negev Desert in the Nahal Boker valley (NB). Our investigations were devoted to the composition of endolichenic fungi inhabiting the thalli of saxicolous lichens covering the rocks at these sites. The studies mainly focused on the effect of various environmental factors on the composition and richness of endolichenic fungal communities. Twenty-eighth lichen species were collected in the selected sites: 13 species in the AZ, 6 species in the NO, and 12 species in the NB. During these studies, 39 endolichenic fungal species were isolated from the lichens collected in the AZ, 60 species from the lichens collected in the NO, and 101 species from the lichens collected in the NB. Fungi occupying the interior of lichen thalli should cope with a variety of harsh environmental factors: restricted nutrient supply, low water availability, limited aeration, and rapid changes in temperature and hydration of the exterior environment. In addition, lichen-associated fungi should withstand the activity of various extracellular secondary metabolites excreted by their host species. Our studies shown the endolichenic communities possessed a set of traits, which helped them to cope with harsh internal and external conditions, including the overall prevalence of melanin-containing fungi with thick-walled and multicellular spores in communities as well as a comparatively high contribution of thermotolerant species and species with meiosporereproducing life cycle stage at the most microclimatically severe habitats. Species richness of the endolichenic fungal communities was associated, to some extent, with the growth form of lichens, being higher in those lichens with thick, warted, and wrinkled thalli and significant lower in species possessing thin and smooth thalli. The lowest species richness of endolichenic communities was registered in the thalli of cyanolichen *Collema cristatum*. This decrease, especially expressed in the summer, was associated with the antifungal effect of substances produced by its cyanobiont, as well as with the heavy dehydration of thalli during the dry season.

Macrofungal biodiversity of the Philipps University Marburg Forest.

Mustafa Isiloglu[1], Robert R. Junker[1], Karl-Heinz Rexer,[1] Hampus Petrén[1]

[1] Evolutionary Ecology of Plants, Department of Biology, Philipps-University Marburg, Karl-von-Frisch-Str. 8, 35043 Marburg, Germany

In this study, the macrofungal biodiversity of Philipps University Marburg forest were investigated by trips carried out in the years of 2021 and 2022. The university forest is located close to village Caldern and 12 km away from Marburg city. The altitude of the forest ranges from 280 m to 412 m. It has an area of 204 ha. with a mixed forest of mainly deciduous trees, dominated with *Fagus* and *Quercus.* After visiting the forest every week, totally 3547 specimens were recorded. These specimens were identified as 434 macrofungal taxa belonging 154 genera and 73 families. These taxa are listed here with photographs of some of them.

Micromycetes in living premises in Minsk city

Tatsiana Shabashova[1], Darya Belomesyatseva[1], Sergei Aleinikav[1]

[1]V.F. Kuprevich Institute of Experimental Botany, Minsk, Belarus, shtgby@gmail.com

As is generally known, due to the presence of a wide range of enzymes, fungi can use various materials of both natural and artificial origin as a food substrate: wood, wallpaper, textiles, oil and water-based paints, plaster, whitewash, cement. The presence of fungi (spores, fragments of mycelium) and their metabolites in the air can have a toxic effect, promote the development of mycoses, and provoke the development of allergic reactions.

We examined more than 30 residential premises in the city of Minsk with signs of mold damage in the form of raids on the walls, ceiling, floor, windows. The isolation of fungi from samples (fragments of wallpaper, scrapings from surfaces) on an agar nutrient medium was carried out in accordance with generally accepted standard methods. In total, 58 species of filamentous fungi were isolated, the predominant number of species belonged to ascomycetes in the anamorphic stage, 3 species in the teleomorph stage, 4 zygomycetes and 2 species of yeast.

When examining residential premises, it was noted that the species composition and structure of mushrooms depend on the time the house was built, the materials used in their construction. In new panel-type houses, where the end walls and joints are most often wet, favorable conditions are created for their colonization by fungi, mainly *Alternaria*, *Cladosporium* and *Penicillium*, and it is noted that these genera are the “pioneers” of settlement, over time they are added and other fungal genera *Acremonium*, *Aspergillus*, *Stachybotrys* and *Ulocladium*. The greatest "species diversity" is observed in houses built in 1960-1980. Regardless of the time of construction of the house, if plastic windows are not properly installed or used, fungi of the genera *Alternaria*, *Cladosporium* and *Ulocladium* settle on them, leading to significant damage to rubber insulation and the surface of the frames. In bathrooms, in addition to the main genera of fungi, hygrophilic fungi of the genera *Acremonium, Humicola*, *Rhizopus* and *Trichoderma* are often found.

An analysis of the structure of the fungal complex shows that the species diversity is dominated by the genera *Aspergillus* and *Penicillium*, but by the degree of dominance and occurrence in the mycobiota of the premises, the representatives of the genera *Cladosporium* and *Alternaria* are the main ones.

MICROPLANTALP Microorganism-Plant Interactions in Alpine ecosystem: a hotspot for studying the impact of climate change.

Giorgia Cavallini[1], Laura Zucconi[1], Angela Augusti[3], Alessandro Bernetti[4], Luigimaria Borruso[2], Enrico Brugnoli[3], Fabiana Canini[1], Federica D'Alò[3], Ilaria Fracasso[2], Olga Gavrichkova[3], Leonardo Montagnani[2], Gianmarco Mugnai[4], Irina Pinchuk[4], Ciro Sannino[4], Maurizio Sarti[3], Mimmo Tanja[2], Benedetta Turchetti[4],

[1]Department of Ecological and Biological Sciences, Tuscia University, Largo dell'Università s.n.c. Viterbo, Italy, giorgia.cavallini@unitus.it, zucconi@unitus.it, canini.fabiana@unitus.it; [2]Faculty of Agricultural, Environmental and Food Sciences, Free University of Bozen-Bolzano, Bozen-Bolzano, Italy; [3]CNR – Research Institute on Terrestrial Ecosystems (CNR-IRET), Porano (TR), Italy; [4]Department of Agricultural, Food and Environmental Sciences, University of Perugia, Perugia, Italy.

Alpine regions exhibit significant environmental diversity, with strong altitudinal gradients and a wide range of climatic conditions within a relatively small spatial area, which makes them ideal hotspots for studying the impact of climate change on soil ecosystems. In particular, while it has been observed that plant cover is increasing at higher altitudes, its consequences on the diversity, abundance, and functionality of associated microbial communities remain uncertain. Prokaryotic and eukaryotic microorganisms living in these habitats play a crucial role in regulating soil health, nutrient cycling, and directly influencing plant growth, development, and resilience. Indeed, one of the most dramatic consequences of climate change is the alteration of the delicate balance between photosynthetic carbon assimilation and its subsequent release into the atmosphere, through respiration. Understanding the intricate ecological processes underlying the interactions between microbial communities and above-ground plant coverage requires an integrated multidisciplinary approach that brings together various fields of expertise. The MICROPLANTALP project, through a study that considers both soil microorganisms and plants, aims at gaining insights into ecological responses under possible global warming scenarios. This will be achieved through the comparison of two complementary transplantation approaches with the aim to characterize the responses of an alpine ecosystem (Mont Blanc area, Val Veny, Courmayeur). Soil monoliths, including native plants, collected at high altitudes (2500 m), are the model on which the study is focused. In the first approach, monoliths were transplanted to a lower latitude (1,000 meters elevation difference), while in the second experiment, other monoliths collected in the same area have been exposed to two different future climate change scenarios at the Montpellier Ecotron facility. Soil bacterial and fungal diversity of samples collected in their natural environment (control samples) and those from the two transplantation experiments will be characterized using a DNA metabarcoding approach (Illumina MiSeq). Additionally, the metabolic activity of microbial communities in soil samples will be evaluated through enzymatic activity assays targeting enzymes involved in C and N cycling. Moreover, the abundance of specific taxonomic groups of interest and of genes involved in the pathways mentioned above will be analysed through qPCR approaches. The data obtained will be compared and integrated with plant growth and carbon fluxes and several soil physicochemical parameters. By analyzing changes in the taxonomic structure and metabolic activities of microbial communities and foresee the plant-soil carbon fluxes under various climatic scenarios, models on how these ecosystems may evolve in the future will be developed. These predictive capabilities are vital for devising effective conservation and management strategies to safeguard these delicate ecosystems from the impacts of climate change.

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Molecular exploration of the diversity within the *Amidella* clade (*Amanita* Pers. : *Amanitaceae* : *Basidiomycota*).

Paulo Oliveira[1], Ricardo Arraiano-Castilho[2]

[1]CIBIO/BIOPOLIS, Address, and MED, Address, Portugal, oliveira@uevora.pt; [2]Department of Ecology and Evolution - University of Lausanne, Biophore - CH-1015 Lausanne, Switzerland. ricardo.arraianocastilho@unil.ch

Species determination in the *Amidella* clade is notoriously difficult, because of the relative dearth of field characters (1) and the rather common occurrence of homoplasies (2). One of the consequences of this is a gross underestimation of the number of species, with a substantial amount of misnamed and unnamed collections presumed to belong to this clade. In order to assess the diversity that should be considered as part of *Amidella*, we retrieved DNA sequences available for this group on nucleotide databases, using a combination of approaches to sample over the entire scope of this group. For the nuclear internal transcribed spacer (ITS) region, a total of 191 sequences were obtained, most of them associated with collected basidiomes, and only a few from ectomycorrhizas or soil samples. Phylogenetic analysis based on the aligned ITS sequences suggests at least 45 clades at species level, of which only up to 12 can be assigned a valid name, 4 more corresponding to provisional names listed in the amanitaceae.org website, and 7 others to *cryptonomina temporaria* on the same resource. It is likely that some of the remaining clades will match named species (as DNA sequences are obtained from available type materials), but it seems likely, too, that other clades are still for “dark taxa”. An ITS-based phylogenetic arrangement is proposed as a framework for studying character evolution within *Amidella* and for designing clade- specific molecular markers to assist in identification (2). It is consistent with the results from the analysis of other molecular markers for the same specimens (nLSU, RPB2, TEF1, B-TUB), and suggests four major branches (Figure 1): one with *Amanita ponderosa* Malençon & R. Heim and allies, another with *A. curtipes* E.-J. Gilbert and allies, a third one including, presumably, the type species *A. volvata* (Peck) Lloyd, and a fourth one presumably containing *A. clarisquamosa* (S. Imai) S. Imai and allies. This study also provides a phylogeny-arranged picture of the worldwide distribution of presumptive *Amidella* species. As expected, collections are predominately from Europe and the Mediterranean, North America and East Asia, but also from Subsaharan Africa and Australia.

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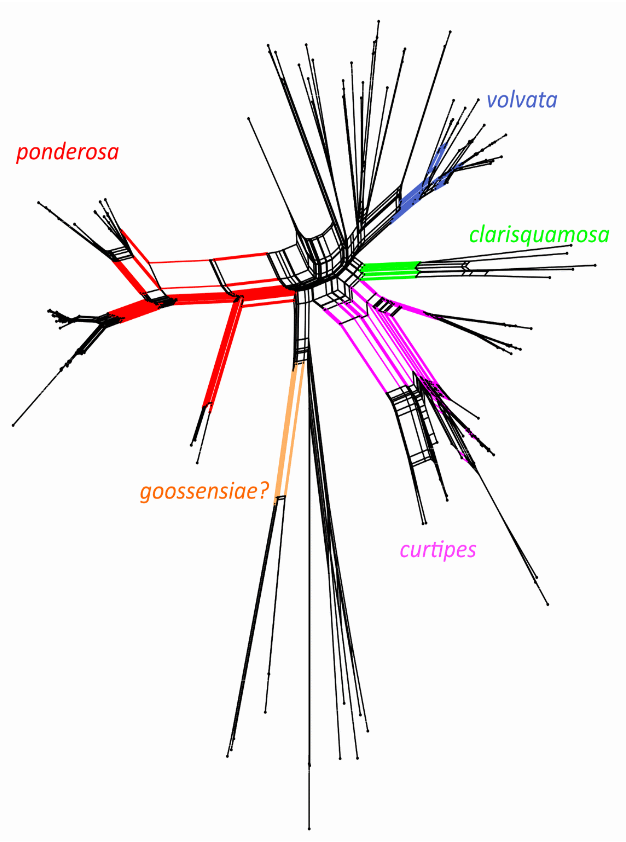

Figure 1. Phylogenetic arrangement of the ITS sequences compiled in the present study, highlighting the branches mentioned in the text, as well as a subbranch ('goossensiae?') of the outgroup.

Molecular identification and genetic characteristics of respiratory microsporidiosis in patients with lung cancer.

Ozlem Ulusan Bagci[1], Can Muftuoglu[2], Filiz Guldaval[3], Ufuk Mert[4], Ayse Caner[2,5]

[1]Ataturk Training and Research Hospital, Microbiology Laboratory, Izmir Katip Celebi University, Izmir, Turkey, drozlemulusan@gmail.com.
[2]Department of Basic Oncology, Institute of Health Sciences, Ege University, Izmir, Turkey, canmuftuoglu1990@gmail.com.
[3] Chest Disease Department, Izmir Dr. Suat Seren Chest Disease and Surgery Training and Research Hospital, Izmir, Turkey, filizguldaval@yahoo.com
[4]Ataturk Health Care Vocational School, Ege University, Izmir, Turkey, ufukmrt@gmail.com.
[5] Department of Bionformatics, Institute of Health Sciences, Ege University, Izmir, Turkey.ayecaner@gmail.com

Lung cancer (LC) is the leading cause of cancer-related deaths in the world (1). The patients with LC are more prone to infections than the normal population due to cancer itself (disruption of organ integrity), treatment managements, and alteration in the immune system (2,3). Infections and their complications are still among the most important causes of morbidity and mortality in cancer patients; however, the diagnosis of infections can be complicated by a scarcity of symptoms or an atypical clinic. Therefore, the diagnosis of pathogens, including opportunistic or neglected microorganisms, is important in terms of reducing mortality. Microsporidia, which are opportunistic fungi, primarily localize to the intestine by ingestion but can disseminate to the respiratory tract or can be acquired by spore inhalation (4). Cancer patients are at higher risk for microsporidia, a life-threatening infection, than the normal population (5,6). We aimed to characterize the prevalence of microsporidia infection for the first time by evaluating the respiratory tracts of patients with LC. In this study, we investigated 98 patients with LC and 103 healthy individuals for microsporidia infection and evaluated the clinical findings of patients who were found to be positive. Sputum and saliva samples were tested by microscopic examination, in addition to pan-microsporidia and genus-specific polymerase chain reactions (PCR). Eight patients with LC had positive results for microsporidia (8.2%), which was significantly higher than the rate in healthy individuals ($P < 0.01$), and they had serious clinical findings. Among these positive patients, PCR revealed microsporidia in the sputum samples of seven patients, and both the sputum and saliva samples of one patient. *Encephalitozoon cuniculi* was identified as the predominant pathogen in 87.5% (7/8) of positive samples. Microsporidia infection was significantly associated with advanced stages of cancer. However, Microsporidia was not detected in the samples of the control group. In conclusion, the study showed that microsporidia infection is a significant risk for cancer patients, with the observed high prevalence. More importantly, respiratory microsporidiosis, whose symptoms often resemble those of other infective agents, should also be considered in cancer patients. Among Microsporidia, especially, *E. cuniculi*, as a respiratory tract infection, should be screened in respiratory samples of cancer patients when they have pulmonary symptoms (The project was funded by TUBITAK with the number 120N924).

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Fungal conservation: Resolving gaps in the distribution of grassland fungi – where are they?

Susan Jarvis[1], Aileen Baird[2], Sean Cooch[2], Matt Wainhouse[2]

[1] UKCEH, Wallingford, UK
[2] Natural England, Bristol, UK, matthew.wainhouse@naturalengland.org.uk

The Grassland Fungi (CHEGD) assemblage represent a functional group of soil saprotrophs dependant on long-established and undisturbed grassland. The UK is of international importance for its grassland fungi assemblages hosting 23 globally threatened taxa (with 11 unpublished VU taxa). These grasslands face an existential threat from changes in land-use and management.

Important grassland fungi sites can be difficult to identify and do not correspond to botanically rich sites. Many are found in the undisturbed, botanically poor, sheep grazed grasslands of the uplands and upland fringe. This makes them especially vulnerable to government-led tree-planting schemes which target such unproductive farmland. Tree planting policy aims to avoid conflict between afforestation and other features of nature conservation value, but this is dependent on high quality data tools to inform decisions. Biological records are useful, but there are significant gaps in their distribution suggesting that a high proportion of the grassland fungi resource is not known about.

It is however possible to use these biological records to model where grassland fungi sites might be located. Here we present the initial outputs from our grassland fungi predictive model for the UK. Using more than >65,000 grassland fungi records, individual species distribution models were constructed for all high priority species with sufficient data and assessed for recording bias. These were combined with environmental variables. Predictions of high value grassland fungi assemblages were made for all locations in England and sites identified where there is a high probability of supporting assemblages that would meet the criteria for national protection. Model validation through eDNA and fruitbody surveys are planned for autumn 2023.

Mycological diversity of Belchishta Wetland – the unique flooded alder forest in Macedonia.

Slavica Tofilovska[1,2], Kristijan Jakimovski[2], Mitko Karadelev[1,2], Katerina Rusevska[1,2]

[1]Institute of Biology, Faculty of Natural Sciences and Mathematics, Ss. Cyril and Methodius University,
Arhimedova 5, Skopje, Macedonia, krusevska@pmf.ukim.mk
[2]Macedonian Mycological Society, Arhimedova 5, Skopje, Macedonia

Belchishta Wetland is probably the best conserved and most important wetland in the country. It is fed by more than 10 karst springs and harbors different types of habitats, one of which is the flooded alder forest that’s the only locality of this habitat in Macedonia. It is found at the foot of Ilinska Planina, at an altitude of about 760 m and it covers an area of 401 hectares, out of which around 60 hectares are under alder forest. The wetland is habitat for numerous endemic and rare species, it is regarded as biodiversity hotspot of high natural value, therefore if was proclaimed as Nature Park (Category IV) according to the Law for Nature Protection that is following the International Union for Conservation (IUCN) categorization. Mycological surveys were conducted in Belchishta Wetland with special focus on the diversity in the flooded alder forest, as well as in the adjacent oak forest and wet meadows. The material was collected in several field research activities during October in 2014 and 2020. The material was identified following the morphological species concept and latest taxonomical knowledge, and it was deposited in the Macedonian Collection of Fungi (MCF). A total of 83 species of fungi have been registered, the majority belong to the phylum Basidiomycota (74), in Ascomycota 8 species and only one species belongs to Myxomycota. The phylum Basidiomycota is represented by 37 families, out of which with highest number of species are: *Mycenaceae* (8), *Polyporaceae* (7), *Marasmiaceae* (7), *Agaricaceae* (5), *Tricholomataceae* (5). Most of the species are lignicolous (52), terricolous are 30 species and one is bryophyllous. Lignicolous specimens were found on the following substrates: fallen alder branch, fallen alder fruit, alder trunk, oak trunk and on living tree of alder. During the research four species were recorded for the first time for the country, *Cortinarius bibulous, Entoloma euchroum*, *Marasmiellus confluens* and *Xeromphalina campanella*, all of them mycorrhizal with *Alnus glutinosa.* The rare species *Lactarius lilacinus* and *Alnicola escharioides* were found, for last this is the second locality in the country. Also, two species that are listed in the Macedonian Red List of Fungi were identified, *Gyrodon lividus* and *Lactarius omphaliformis* which are categorized as endangered. *Gyrodon lividus* is found at eight sites in the country, while *Lactarius omphaliformis* only in two sites. With this research for the first time a systematized data on the fungi of Belchishta Wetland is presented. The diversity of fungi is probably higher, and more detailed research is necessary, especially in the spring and summer period in order to get a more complete list of species richness. Taking all this into account, it can be concluded that Belchishta Wetland is an important area for the diversity of fungi.

Novel anticoagulant enzyme produced by *Aspergillus tabacinus*.

Viktoriia Lavrenova[1], Alexander Osmolovskiy[2]

[1]Lomonosov Moscow State University, Department of Biology, 119234 Moscow Leninskiye Gory 1b12, Russia, pkviktoria@mail.ru; [2] Lomonosov Moscow State University, Department of Biology, 119234 Moscow Leninskiye Gory 1b12, Russia, aosmol@mail.ru

Cardiovascular diseases have been the leading cause of death worldwide for more than a decade. The vast majority of these diseases is associated with thrombosis, besides thrombosis can be an independent ailment as well as an accompanying factor for other illnesses. All these considerations make the search for new hemostasis regulating drugs an urgent task for both fundamental science and pharmacology. Proteases of microscopic fungi including the species of *Aspergillus* genus are promising candidates for the development of new anticoagulant and thrombolytic drugs (1). Here we identified a new potent anticoagulant enzyme from the marine-derived strain *Aspergillus tabacinus* BEOFB3260m. The micromycete was cultivated in two stages. Firstly, fungal spores were washed off from a 7-day old slant and transferred to the seed medium of the following composition, % (w/v): wort – 6.7, glucose – 1.0, peptone – 0.1, pH 5.5–6.0. Secondly, after 48 hours of cultivation at 28˚C and 200 rpm 3% (v/v) of the seed medium were transferred to fermentation medium №1 (% (w/v): glycerol - 7.0, glucose - 3.0, fish flour hydrolysate - 3.0, NaNO3 - 0.2, MgSO4 - 0.1) or fermentation medium №2 (% (w/v): starch - 1.0, glucose - 3.5, fish flour hydrolysate - 0.5, peptone - 0.5, MgSO4 - 0.05, KH2PO4 - 0.05, NaCl - 0.2) and cultivated under different conditions until maximum proteolytic actvity of the culture liquid was reached. Maximum protein C-like activity (measured spectrophotometrically with chromogenic substrate pGlu-Pro-Arg-pNA) was observed under 7-day cultivation in fermentation medium №2, pH 7.0-8.0, 28˚C. The enzyme was isolated from the described culture liquid through precipitation with ammonium sulfate at 80% saturation followed by dialysis of the precipitate against 0,05 M phosphate buffer, pH 8.2, and subsequent lyophilization. Further purification was performed by column isoelectrofocusing, but Laemmli electrophoresis showed that fraction with high protein C-like activity consisted of several proteins. However, the proteolytic enzyme presented as one band with molecular mass of approximately 30 kDa on casein zymogram. Detection of glycosylated proteins on gels (2) and inhibitory analysis demonstrated that the investigated enzyme is a non-glycosylated serine protease which is inhibited by soybean tripsin inhibitor, but is not inhibited by universal serine proteases inhibitor PMSF. The proteolytic activity of *A. tabacinus* secreted protease towards substrates of thrombin (Tos-Gly-Pro-Arg-pNa), factor Xa (Ac-Leu-Gly-Arg-pNa and Z-D-Arg-Gly-Arg-pNa), plasmin (H-D-Val-Leu-Lys-pNA) and protein C (pGlu-Pro-Arg-pNa) exceded proteolytic activity of anticoagulant enzymes from other *Aspergillus* species (3), what makes *A. tabacinus* a perspective producer of anticoagulant protease for pharmacology.

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Mycobiota of the soil just below the base of fruiting body of eight wild mushrooms collected in Korea.

Seong Hwan Kim[1], In Hee Jung[2], Na Young Yoon[3], Pyung Yeol Ko[4], Hye Sung Park[5]

[1]Dankook University, Cheonan, Republic of Korea, piceae@dankook.ac.kr; [2]Dankook University, Cheonan, Republic of Korea, hosi4400@naver.com; [3]Dankook University, Cheonan, Republic of Korea, yummna3454@naver.com; [4]Agriculture Research Institute, Jeju National University, Jeju, Republic of Korea; [5]Mushroom Research Division, National Institute of Horticultural & Herbal Science, Eumseong, Republic of Korea

Rhizosphere soils of 8 species of wild mushrooms were analyzed to obtain ecological information on the rhizosphere of wild mushrooms in Korea. Soil samples of mushroom rhizosphere were collected from *Amanita hemibapha*, *Cudonia circinans* on Jirisan Mountain, from *Macrocybe gigantea*, *Armillaria mellea* in Jeju-si, and from *Amanita rubescens*, *Morchella esculenta*, *Pseudotulostoma japonicum*, *Tricholoma bakamatsutake* in Seogwipo-si. The results of analyzing the physical and chemical characteristics of the rhizosphere soils of mushrooms revealed that *A. hemibapha*, *A. mellea*, and *A. rubescens* were from sandy loamy soil, while *C. circinans*, *M. gigantea*, and *P. japonicum* were from loamy sand soil, and *M. esculenta* was sandy soil. Soil analysis was not possible for *T. bakamatsutak* because of its low soil composition and high organic matter content. Soil acidity was close to acidity in the rhizosphere of *M. gigantea* and slightly acidic in the case of the rhizosphere of the other seven species of mushrooms. There was a difference in the species diversity of fungi in the results of the culture and non-culture methods. A total of 27 fungal species in 14 genera were isolated and identified. The genus *Trichoderma* was dominant followed by *Penicillium*, *Umbelopsis*, and *Mortierella*. As a result of analyzing the species richness of fungi present in the rhizosphere soils of eight wild mushrooms, the species richness was high in the rhizosphere soils of *M. esculenta* collected in Seogwipo-si. Species diversity showed a value of 1 or more in the rhizosphere soil of *P. japonicum* and rhizosphere soil of Mushroom oyster mushroom, and a value of less than 1 in the other rhizosphere soils. In order to compare the community morphology of the rhizosphere soils of 8 species of wild mushrooms, the community morphology was investigated by PCoA analysis. PCoA 1,2 (32.18%, 23.11) results showed the fungal communities were similar to each other, except for the rhizosphere soils of the wild mushrooms, *C. circinans* and *P. Japonicum*. As a result of mycobiota analysis conducted by a non-culture method, the genus *Mortierella* was dominant in the rhizosphere of *A. mellea*, *M. esculenta*, and *A. rubescens*. According to the Simpson and Shannon index, fungal diversity was the highest in rhizosphere soil of *M. esculenta*. Our data provide a basis for understanding the ecological interactions between mushroom and its rhizosphere fungi.

Searching for the rare fungus *Flaviporus citrinellus*.

Reda Iršėnaitė[1], Saulis Skuja[2],

[1]Nature Research Centre, Žaliųjų ežerų Str. 47, LT-12200 Vilnius, Lithuania, reda.irsenaite@gamtc.lt
[2]Vilnius University, Life Science Center, Institute of Biosciences, Saulėtekio ave. 7, LT-10257 Vilnius, Lithuania, saulis.skuja@gf.vu.lt

Lemon-coloured Antrodiella (*Flaviporus citrinellus*; syn. *Antrodiella citrinella*) - lemon yellow wood polypore prefers old spruce (*Picea abies*) forests with plenty of coarse wood debris and moist microclime. The fruiting body appears in early spring and late summer on spruce logs that was previously decomposed by the perennial fruiting of *Fomitopsis pinicola*, which develops over many years from spring to autumn (1). It is distributed only in Europe, mainly in the central and northern part and globaly red listed as endangered (EN) (2).

F. citrinellus was collected for the first time in Lithuania in the Curonian Spit, in a rather isolated area, in a small fragmented remnant of an old spruce forest on a steep slope near a stream. For the study sites were selected with spruce trees over 80 years old, with amounts of woody debris exceeding 5 cubic metres per hectare and situated close to habitats with excess moisture. The aim was to: 1) assess the habitat requirements of the species; 2) highlight its distribution and rarity status; 3) compare the efforts of experts and citizen scientists in the search for rare species.

Of the 30 sites checked, 15 were found to contain fruiting bodies of *F. citrinellus*. The majority of records were obtained from moderately decomposed fallen spruce stumps with an average diameter of 59.6 (9.3SD) cm and fallen trunks with an average diameter of 48.3 (4.3SD) cm. Other rare species such as *Gyromitra sphaerospora, Junghuhnia collabens, Phellinidium ferrugineofuscum, Phlebia centrifuga, Rhodofomes roseus*, which are typical of spruce coarse woody debris, were recorded on the same or neighbouring logs.

Differences between sites with and without *F. citrinellus* fruiting bodies were only present at higher *Picea abies* ages (MRPP, A=0.141, $p<0.01$) in the habitat. Site size, age of the dominant tree on the site, and humidity did not differ significantly.

27 sites were selected and screened without knowing the presence of *F. citrinella*. Of these, 12 were found (44%). Information on three sites was provided by trained ecologists and no new information on the sites was found in the iNaturalist citizen science database.

In general, our results show that, of all the environmental factors assessed, the age of spruce in the stand was the most important for this species. Targeted specialist searches have revealed many new occurrences of this rare species, with training of environmental specialists contributing to this, while citizen science has contributed little to the discovery of this species. The criterion of global threat to the species needs to be assessed on the basis of new data, including from other countries.

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Species diversity of microfungi in common medicinal herbs in Slovakia.

Katarína Pastirčáková[1], Martin Pastirčák[2]

[1]Slovak Academy of Sciences, Institute of Forest Ecology, Department of Plant Pathology and Mycology, Akademická 2, SK-94901 Nitra, Slovakia, uefezima@hotmail.com; [2]National Agricultural and Food Centre, Plant Production Research Institute, Bratislavská 122, SK-92168 Piešťany, Slovakia, martin.pastircak@nppc.sk

Medicinal plants have a tremendous potential as both natural and genetic resources. In Slovakia, however, research on diseases and their causal fungi on medicinal plants has not been conducted sufficiently. Thus, a survey on diseases of wild and cultivated medicinal plants was organized. Two plants, namely *Achillea millefolium* and *Hypericum perforatum*, were chosen for their widespread use in traditional medicine and extensive native range in Slovakia. In the survey since 2019, over 80 diseased plant samples were collected from a total of 40 locations in different distinct in Slovakia. The classical method of isolation and morphological analyses were used to identify fungal communities on symptomatic plants. In dead parts of stems, fungi were identified by microscopic analyses on base of morphological characteristics of the fruiting bodies and spores. In 48 samples of *Achillea millefolium* plants, a total of 86 fungal isolates representing 22 genera, including dark septate and sterile fungi, were identified. *Phomopsis* (83%), *Septoria* (26%), and *Camarosporium* (17%) were the most frequent fungi found in necrotic lesions on *A. millefolium* stems. Other frequently isolated fungi included *Alternaria*, *Colletotrichum*, *Dichomera*, *Fusarium*, *Phoma*, and *Stemphylium*. *Golovinomyces macrocarpus* causing powdery mildew disease on yarrow leaves has been recorded rarely. In 36 samples of *Hypericum perforatum* plants, a total of 97 fungal isolates representing 24 genera were isolated. The most frequent fungi detected in necrotic lesions on *H. perforatum* stems were *Diploceras* (47.2%), *Septoria* (25%), and *Mycosphaerella* (17%). Other frequently isolated fungi included *Ascochyta*, *Botrytis*, *Clathrospora*, *Colletotrichum*, *Coniothyrium*, *Fusarium*, *Melanconium*, *Microdiplodia*, *Phaeosphaeria*, *Phoma*, *Phomopsis*, *Pleospora*, *Scopinella*, and *Trichometasphaeria*. The symptoms of powdery mildew (effuse-to-dense white patches of mycelium, primarily on adaxial leaf surfaces) caused by fungus *Erysiphe hyperici* have been recorded very often. The study on fungal diseases of medicinal plants is important for conservation and stable production of such useful plants. The research of fungal diversity on other medicinal plant species should also be focused in the future. The present study showed the diversified fungal flora on medicinal plants in Slovakia.

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Strong impact of microclimatic, climatic, and anthropogenic factors on cultivable soil microfungal communities at a canyon in the Mediterranean region.

Isabella Grishkan[1], Shmuel Raz[2], Eviatar Nevo[3]

[1]Institute of Evolution, University of Haifa, 199 Aba Khoushy Ave, Mount Carmel, Haifa 3498838, Israel, bella@evo.haifa.ac.il;
[2]Department of Information Systems, University of Haifa, 199 Aba Khoushy Ave, Mount Carmel, Haifa 3498838, Israel, razshmu@gmail.com
[3]Institute of Evolution, University of Haifa, 199 Aba Khoushy Ave, Mount Carmel, Haifa 3498838, Israel, nevo@evo.haifa.ac.il

One of the most interesting and intriguing aspects of biodiversity exploration is testing the effect of microscale environmental variability on biodiversity patterns. The Institute of Evolution, University of Haifa, Israel, launched such microscale test in the framework of the "Evolution Canyon" (EC) biodiversity research program (1). Following the program, we studied the effect of microclimatic, climatic, and anthropogenic factors on soil cultivable microfungal communities at one of the ECs - Wadi Metzar, located at the Golan Heights. The canyon consists of the mesic temperate north-facing slope (NFS) and the more xeric south-facing slope (SFS). The contrasting microclimatic conditions on the opposite slopes are associated with differences in solar radiation and accompanied by differences in plant communities covering these slopes. The main goals of the study were as follows: (i) to examine the effect of microscale environmental variability on the composition and structure of soil microfungal communities by comparing the communities from the NFS and SFS, (ii) to assess the mycological consequences of agricultural usage of the area by comparing the communities in the currently recovered and previously disturbed soil of SFS, and (iii) to reveal the possible impact of global warming on soil mycobiota by comparing the current and previous (17 years before) natural communities from the NFS. By means of the soil dilution plate method, a mycobiota accounting for 94 species from 47 genera was isolated. The communities' composition was subjected to the pronounced spatial (interslope) and seasonal (summer-winter) variations. While xerotolerant melanin-containing species and thermotolerant *Aspergillus* spp. predominated in the soil of SFS, peaking up in the summer, mesophilic *Penicillium* spp. were especially abundant at the NFS. The quantitative parameter – density of microfungal isolates, exhibited the strongest seasonal variations being more sensitive to the fluctuation in soil temperature and moisture. Comparison of microfungal communities in the currently recovered and previously disturbed soil at the SFS showed that intensive pasturage followed by soil degradation led to the simplification of communities, decreasing their diversity level, and caused the prevalence of species with different ecological preferences. Whereas stress-tolerant aspergilli and melanin-containing species prevailed in the native communities of SFS, the mesophilic *Penicillium aurantiogriseum* was dominant in the disturbed soil of the slope, accompanied by the very fast-growing mycoparasitic *Trichoderma koningii*. Comparison of the NFS microfungal communities isolated from the native soil in the summer 2002 and 2019 revealed changes in the community structure, which could be related to global warming: a substantial decrease in the abundance of mesophilic *Penicillium* spp., accompanied by a remarkable increase in the contribution of thermotolerant aspergilli and xerotolerant melanin-containing species. These changes were associated with the stress-resistant fungal traits, which might be useful under increasing soil temperature and desiccation.

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The diversity and distribution of indoor fungal communities.

J. Oja1, F. Shafiq, S. Anslan, L. Tedersoo

[1]*Department of Botany, University of Tartu, J. Liivi 2, 50409 Tartu, Estonia*

Indoor fungal communities play an important role in our daily lives as they can have adverse effects on both occupants' health and the health of wooden constructions. We are mostly aware of the toxic black mold Stachybotrys chartarum, which can cause harm to our health, and the wood rotting fungus *Serpula lacrymans*, which can destroy our wooden buildings. However, the diversity of indoor fungi is much higher than expected and highly diverse communities are expected to do us good than harm. Our objective was to identify dust-associated fungi in homes across the world and characterize factors that shape their distribution by using citizen scientists' help and high-throughput sequencing technologies. Our dataset is the largest yet assembled for indoor fungal communities, encompassing samples from six continents. Our findings shed new light on the global distribution of indoor unnoticeable organisms, and we show that several of them have a unique distribution pattern. In addition, we reveal different factors affecting dust-associated fungal communities. In sum, we get a better understanding of the environment around us and how it may impact our well-being in buildings.

The Italian Fungi Census Initiative.

Francesca Floccia[1]

[1]Italian Institute for Environmental Protection and Research (ISPRA), Rome, Italy

In 2022 the Network for the study of mycological diversity of ISPRA (the Italian Institute for Environmental Protection and Research) promoted a national Fungi census initiative, the first one in Italy.

The first objective of the initiative is the development of a national database, available to the public, of macromycetes present in the habitats of the territory.

The Italian Fungi census initiative is carried out with collection of both field mycological data and fungal samples, sent to ISPRA by the contributors who adhere to the initiative.

The data are collected and processed by ISPRA and disseminated in an opened information system called Fungi Information System (SIF).

In the same way, samples are collected by contributors and sent to the ISPRA fungarium both for conservation and molecular analyses.

Each contributor is owner and responsible for the data sent to ISPRA.

All fungal species have the same importance because it is a census activity!

From the census activity it is possible to extrapolate the following data:

- check-list of Italian fungal species matched with habitats
- list of rare or threatened species
- indicators of habitat quality and climate change.

Specific monitoring will be carried out for some species and habitats.

The census initiative is coordinated by the Network for the study of mycological diversity which is a system that integrates human and technological resources.

The Network is composed by:

- ISPRA – National Institute for Environmental Protection and Research
- Mycological groups, mycologists and experts in mycology
- Scientific committee, that defines standards, activities, themes, and orientation of scientific initiatives.

To the census initiative can participate:

- MYCOLOGIST listed on the national registry.
- EXPERTS IN MYCOLOGY, who can join the Network by sending their curriculum vitae which will be examined for the assessment of skills.

Field and historical data must be sent via ISPRA apps (mobile and web).

The role of *Verticillium dahliae* in the dieback of *Ailanthus altissima* in Trentino - South Tyrol.

Claudia Maria Longa[1], Maria Claudia Ferretti[2], Giorgio Maresi[2]

[1]Fondazione Edmund Mach – Research and Innovation Centre, Via E. Mach 1, 38010 San Michele all'Adige, TN, Italy, claudia.longa@fmach.it; [2]Fondazione Edmund Mach – Technology Transfer Centre, Via E. Mach 1, San Michele A.A. (TN), Italy, giorgio.maresi@fmach.it.

Ailanthus altissima (Miller) Swingle, known as Tree-of-heaven, is a widespread invasive plant species intentionally introduced in Europe as an ornamental and shade tree. It is now widely naturalized and occurs throughout the warmer climatic zones, which has become the most dominant tree species along the edges of forests and roadways. The negative implications of *A. altissima* establishment include the replace of the indigenous plant communities associated with changes on the soil properties and on the composition of the litter fauna. Its eradication is difficult, as it has a high regenerative and reproductive capacity. For these characters, ailanthus has been included in the list of invasive alien species of European Union concern in 2019, so obliging European countries to do monitoring and active control to check its spread. A destructive naturally occurring Verticillium wilt disease represents a potential control of *A. altissima* populations. The causal agent of the disease was identified as *Verticillium nonalfalfae* Inderb. et al. formerly *V. albo-atrum* Reinke & Berthold and *V. dahliae* Kleb in the USA, Austria and Spain. Whereas only *Verticillium dahliae* were isolated in Hungary and in Italy. In Trentino South Tyrol, Ailanthus stand ranging from regeneration to canopy trees displayed foliage wilting, vascular discoloration, and epicormical shoots on affected stems, followed by tree death were observed since 2017. *V. dahliae* was identified as the only causal agent (1) and the disease incidence has been monitored in the last six years to evaluate the potential of *V. dahliae* to control Ailanthus invasion in urban areas and natural environments. A field work was carried out in the whole range of ailanthus colonization trying to individuate the affected stands: 171 points with declining plants were observed with several already dead trees, with a constant increase of the foci during the years. The presence of *V. dahliae* was detected by isolation on media and using real-time qPCR approach with specific primer pair VertBtF/VertBt-R (2). *V. dahliae* was identified as associated with the decline in most of the affected stands (119), while in the other points samples were too old to permit isolation and molecular texts or otherwise affected plants were not approachable. The disease was present in almost all the valleys of the region, either in wood condition or in urban contest. Pathogenicity test confirmed the virulence of the obtained strains on *A. altissima* seedlings. The consistency of the data on the distribution of *Verticillium* wilt in Trentino-South Tyrol suggests a considerable capacity for the spread of the disease, which appears to be undergoing strong and rapid expansion. The spread of Ailanthus wilt is proving to be a potential natural method to contain the populations of one of the worst invasive plant species in Europa.

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Tolypocladium parasiticum Barron, a polyextremophilic fungus.

Violeta Oro[1], Boris Pisinov[1], Marijenka Tabakovic[2], Zoran Sekulic[3], Nenad Trkulja[1]

[1]Institute for Plant Protection and Environment, Teodora Drajzera 9, Belgrade, Serbia, viooro@yahoo.com;
[2]Maize Research Institute „ Zemun Polje “, Slobodana Bajića 1, Belgrade, Serbia, mtabakovic@mrizp.rs
[3]Institute of Public Health of Belgrade, Bulevar despota Stefana 54a, Belgrade, Serbia, zoran.sekulic@zdravlje.org.rs

In the past decades, great attention was devoted to extremophilic microorganisms. Extremophiles survive in extreme conditions, such as extreme temperatures, high saline, acidic, and alkaline solutions or environments with increased heavy metal content (1). Heavy metals are the most persistent and complex kind of pollutants to remediate in nature. Metals, unlike most organic compounds, do not undergo metabolic degradation and accumulate in living tissues. Among heavy metals and metalloids, Zn, Ni, Co, and Cu are more toxic to plants, whereas As, Cd, Pb, Cr, and Hg are more toxic to higher animals (2). They not only degrade the quality of the atmosphere, water bodies, and food crops, but also threaten the health and well-being of animals and human beings. Lead toxicity affects the liver, kidneys, spleen, and lungs, causing neurotoxicity, especially in children. Cadmium ingested from food can damage the kidneys, liver and bones. Zinc is associated with anemia and tissue pathology, and arsenic accumulates mainly in the liver, kidney, heart, and lungs, but to a lesser extent in muscle and nerve tissue, and it is defined as a carcinogen (3). Most contaminants are of anthropogenic origin, but some contaminants have a natural pedo-geochemical origin. Tolypocladium parasiticum Barron, is a known entomo- and nematode-pathogenic fungus. The fungus is an endophyte of Holcus lanatus (4) and halophilic Salicornia europea (5). It is a member of the family Ophiocordicipitaceae, within the order Hypocreales, the class Sordariomycetes, and the phylum Ascomycota. Tolypocladium parasiticum was found in an abandoned medieval mine near the town of Kursumlija in the southern part of the Republic of Serbia. The fungus was isolated from soil and maintained on PDA at 27°C. Species identity was confirmed by molecular analyses and sequencing, using 2234C and 3126T ITS primers (6). The GenBank accession number OR018820 was obtained for this species. Previous research on the stress resistance investigated fungal cold activity, UV-B tolerance, and wet-heat tolerance (7). Our study reports for the first time that the fungus survives at extremely low pH (3.1), high Zn (204 mg/kg), Pb (74 mg/kg), Cd (0.6 mg/kg) and As (54 mg/kg) contents in soil. Our results also revealed that Tolypocladium parasiticum is a polyextremophile, i.e., adapted to life in habitats where various extreme physicochemical properties occur.

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You can't replace quality with quantity: the role of natural forest continuity (and size) for the occurrence of rare and threatened macrofungi.

Jan Holec[1], Pavel Hubený[2], Tomáš Kučera[3]

[1]Mycological Department, National Museum, Cirkusová 1740, Praha 9, CZ-193 00, Czech Republic, jan.holec@nm.cz; [2]Šumava National Park Administration, 1. máje 260, CZ-385 01 Vimperk, Czech Republic, pavel.hubeny@npsumava.cz; [3]Department of Ecosystems Biology, Faculty of Science, University of South Bohemia, Branišovská 1760, České Budějovice, CZ-370 05, Czech Republic, kucert00@prf.jcu.cz

Three localities of differently affected natural forests of Central Europe composed by *Fagus*, *Picea* and *Abies* were compared in terms of the diversity and species value of macromycetes on fallen *Picea abies* and *Abies alba* trunks. The original virgin forest was represented by the Boubínský prales near Vimperk (Czechia) almost untouched by man (1). Lowest degree of natural forest preservation (2) – the near-natural forest – was represented by the Zámecký les forest near Železná Ruda (Czechia) that has been extensively used by the neighboring glassworks for the past three centuries, but still today looks like a primeval forest. For the category of natural forest affected only by sparse selective logging in the past, we used published data from the nearby Mittelsteighütte reserve (3, 4) near Zwiesel (Germany). For all three localities, we collected historical data on their forestry use. Interestingly, the trunks of both *Picea* and *Abies* were species-richer in Zámecký les than in Boubínský prales (5, 6, 7), especially in the case of *Abies*. However, the situation was completely different when we evaluated the representation of generally rare and/or threatened fungi (8), called species of special interest (SSI). They were highly represented in the Boubínský prales, less so in Mittelsteighütte, and least of all in the Zámecký les forest. As the localities are located in the same mountain range and have very comparable habitat conditions, the only factors responsible for these differences are the degree of human influence and also the size, as the Zámecký les is 4 times smaller than the other two. These results demonstrate three facts: a) the substrate, such as a fallen trunk, is always inhabited by some fungi, regardless of the forest naturalness, b) the representation of rare and/or threatened fungi is strongly dependent on the continuity of the forest, and c) the classification of natural forests (2) on the scale of virgin (primeval, original) forest - natural forest – near-natural forest agrees well with the representation of SSI species. In other words, rare and endangered species of fungi are good indicators of forest continuity and the degree of its preservation (naturalness).

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