

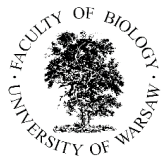
**18<sup>th</sup> Congress  
of European  
Mycologists**



**WARSAW-BIAŁOWIEŻA  
POLAND 16-21.09.2019**

# **Abstract Book**

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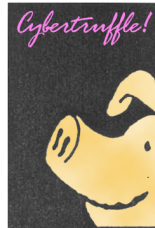
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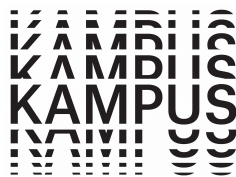
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Edited by Piotr Mleczko

ISBN 978-83-940504-5-0      Polish Mycological Society, Warsaw

Suggested citation:

Name of the first Author of abstract et. al. 2019. Title of abstract. In: Mleczko P. (ed.), Abstract Book, XVIII Congress of European Mycologists, 16-21 September 2019, Warsaw-Białowieża, Poland. Polish Mycological Society, Warsaw, p. xx

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The list of all participants, including the affiliation countries and the e-mail addresses, is presented at the end of the Book. The page number refers to the abstract where the participant is the presenting author.

The texts and the titles of abstracts as well as the names of authors are given in versions submitted during registration. Presenting authors are marked in bold and underlined.

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## OPEN LECTURE

### **Mycology: a recent weapon in the forensic armoury**

**Patricia E.J. Wiltshire**

*University of Southampton, Southampton, UK*

In the past, mycology has contributed to forensic investigation only occasionally, and has been involved mostly in cases of poisoning. In recent years, however, it has been pivotally important in investigating a large number of both civil and criminal cases. When combined with palynology, it is particularly valuable for linking places, objects, and people by providing trace evidence. Mycology can also give information on *post mortem* interval (time since death), and the timing of other events, as well as providing intelligence in the location of clandestine graves and missing persons.

An outline will be given on the ways in which fungi have been used in forensic investigation, and this will be exemplified by case histories showing how fungi have helped in crimes that have resulted in confession and/or conviction.

## KEYNOTE LECTURES



## The evolution of mycorrhizal genomes and transcriptomes

**Annegret Kohler**<sup>1</sup>, Eniko Kiss<sup>2</sup>, Emmanuelle Morin<sup>1</sup>, Shingo Miyauchi<sup>1</sup>, Laszlo G. Nagy<sup>2</sup>,  
Igor Grigoriev<sup>3</sup>, Francis Martin<sup>1</sup>, Mycorrhizal Genome Initiative

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In boreal and temperate forests, trees rely on ectomycorrhizal symbiosis to acquire the limited nutrients in the soil, but the establishment of this mutualistic association leads to dramatic morphogenetic changes in both symbiotic partners. To elucidate the genetic basis and the evolution of the mycorrhizal lifestyle we took advantage of the large fungal genome sequencing programs ongoing at the Joint Genome Institute.

The comparison of 112 genomes from mycorrhizal fungi, wood decayers and litter decomposers revealed several independent lifestyle transitions from saprotrophism to mutualism. A general trend within ectomycorrhizal genomes, but not in ericoid and orchid mycorrhizal genomes, is the reduction of genes coding for plant cell wall degrading enzymes. However, ectomycorrhizal species seem to have retained diverse sets of enzymes depending on their ecological niches. The availability of mycorrhizal genomes enables us as well to compare mycorrhizal transcriptomes and to identify symbiosis-related genes.

We could show that both conserved and clade-specific genes, as for example effector-like mycorrhiza-induced small secreted proteins (MiSSPs), are used by *Laccaria bicolor* to establish symbiosis with the roots of the host tree *Populus*. Using a phylostratigraphy approach we compared the «symbiosistoolbox» of 15 mycorrhizal interactions. Again the age distribution of mycorrhiza-induced genes showed two peaks corresponding to ancestral genes made of conserved gene families and species-specific orphan genes.

About 45% of the mycorrhiza-induced genes predate the origin of mycorrhiza, suggesting gene cooption as a major mode of recruiting genes for symbiosis. Conserved mycorrhiza-induced genes showed little overlaps across species, suggesting that independently evolved mycorrhizal lineages have co-opted different genes. Interestingly, induced genes are coding for the same functions but without orthology, like small-secreted proteins, transporters or carbohydrate active enzymes.

## Discovering the extent of fungal diversity

**David L Hawksworth<sup>1,2</sup>**

<sup>1</sup>*The Natural History Museum, London, UK*

<sup>2</sup>*Royal Botanic Gardens, Kew, UK*

The extent of species diversity amongst the fungi only started to be fully recognized the last decades. The pattern of species description has been accelerating over the last decade with the advent of molecular approaches (and despite the ending of the naming of separate morphs of the same fungus in 2011). There is no indication of numbers discovered falling, and the number of mycologists is the key factor limiting the numbers. But how many species remain to be discovered to achieve a complete global inventory? Prior to the molecular techniques, the focus was on ratios extrapolated from comparisons of the number of plants and the number of fungi from all habitats collected or isolated in a site. A ratio of around 5-6:1 was derived from two particularly well-studied areas in the UK, supplemented by several other independent data sets, and seems to hold also for the tropics. Now, molecular data provide a new dimension to morphospecies-based estimates, in terms of the numbers of cryptic species within long-recognized morphospecies, and environmental sequences. Notwithstanding issues in the interpretation of environmental sequence data, the conclusion that estimates based on visible fungi are far too low is inescapable. Recent work on one soil sample from one well-studied UK wood leads the ratio being increased to 22:1 – and yielded evidence of 132 genera not detected in field surveys. It now emerges that there are probably 2.2-3.8 million species of fungi, suggesting that as many as 97% of the species remain undescribed. How to name species from sequences alone is a challenge we cannot avoid and must resolve at IMC12 in Amsterdam in 2022.

## **Geomycology: metals and minerals, bioremediation and biorecovery**

**Geoffrey Michael Gadd**

*Geomicrobiology Group, School of Life Sciences, University of Dundee, Dundee, DD1 5EH, Scotland, UK*

“Geomycology” is an important part of “geomicrobiology” and can be defined as the impact of fungi on geological processes, including bioweathering of rocks and minerals, metal and metalloid transformations, and element and nutrient cycling. Fungi are important geoactive agents in soil, rock and mineral surface layers, whether free-living or in symbioses with phototrophs, and also significant biodeteriogens in the built environment. Many geomycological processes, dependent on hyphal growth form and chemoorganotrophy, are of relevance to pollutant fate in the environment altering metal mobility through such processes as mineral dissolution, metal accumulation and biomineralization. This presentation will emphasize some important activities of fungal systems in the transformation of metal(loid)s such as Se, Te, Pb, U, Mn, Ca and Co where the formation of elemental forms, or insoluble phosphate, oxide, carbonate or oxalate minerals can provide a means of metal immobilization, not only relevant to bioremediation but also to the biorecovery of valuable elements. Some biometal(loid)s/biominerals are formed at the micro- and nanoscale providing further interest for the development of useful products, including novel electrochemical biomaterials. Finally, mention will be made of the biodeteriorative properties of fungi regarding the destruction of cultural heritage and mineral-based building materials, including concrete, which may have consequences for nuclear decommissioning and radionuclide containment.

## **The Evolutionary Origin and Stability of Cultivation of *Termitomyces* by Termites**

**Duur Aanen**

*Laboratory of Genetics, Plant Sciences Group, Wageningen University, Wageningen, the Netherlands*

The fungus-growing termites cultivate fungi of the basidiomycete genus *Termitomyces*. The symbiotic fungi descend from a single domestication some 30 million years ago in central tropical Africa. The symbiosis is ecologically highly successful and has allowed the colonisation of savannas and several out-of-Africa migrations to Asia. In my talk I will first discuss our current understanding of the evolutionary history of the symbiosis, including links with non-domesticated fungi and their characteristics. Next, I will discuss experimental work on the stability of fungus cultivation. Within a colony, the termites propagate their fungi in large-scale monoculture, via asexual spores inoculated in their faeces. I will discuss in vitro experiments showing the benefits of monoculture cultivation and how monocultures originate from initial mixed cultures when a colony starts. Finally, I will discuss the risks of monoculture cultivation.

## **Comparative Genomics of Smut Fungi to Bridge the Gap between Systematics, Ecology and Function**

**Dominik Begerow**

*AG Geobotanik, Ruhr-Universität Bochum, Bochum, Germany*

Smut fungi evolved in two lineages of Basidiomycota and their systematics is based on ultrastructure of the parasite-host interface and molecular phylogenetic data. Both lineages are dimorphic with parasitic and saprobic stages, host specific and co-evolved with their hosts. Their joint evolution is often characterized by lineage tracking resulting in host specificity and one-to-one relationships, like in other fungal parasitic groups. Therefore, adaptation and specialization to the host plant seems to be crucial and should involve genes or regulatory pathways governing host specificity. To identify genes relevant for host specificity comparative genomics is becoming a relevant method as more and more genomes of fungi are available. Examples from both lineages will be introduced.

In the model system of *Microbotryum* species, we produced artificial hybrids between the two host-specific species *M. lychnidis-dioicae* and *M. silenes-acaulis* and applied strong experimental selection on different host plants to identify genes necessary for successful infections. Genome comparison of the two species revealed that most gene families are shared and the majority of genes are conserved, indicating very similar biological features of both species, including host adaptation and infection processes. Lower nucleotide identity of genes encoding for secreted proteins might indicate their importance for host specific interaction, as it is known from other plant pathogens. Moreover, we identified 211 candidate genes that occur in each hybrid and backcross genome that were posed under host-driven selection and might therefore play a crucial role in host specialization.

In contrast, a broader perspective in Ustilaginomycota allows to identify core genes relevant for general aspects of infection, without any signs of host-specificity. Therefore, the comparison of the two independent smut lineages allows to understand ecological and evolutionary constraints of smut fungi.

## Time to re-think fungal ecological niches? Endophytic abilities in ectomycorrhizal taxa

**Marc-André Selosse**<sup>1,2</sup>, Laure Schneider-Maunoury<sup>1</sup>, Florent Martos<sup>1</sup>, Julita Minasiewicz<sup>2</sup>

<sup>1</sup>*Institut de Systématique, Evolution, Biodiversité, Muséum national d'Histoire naturelle, Paris, France*

<sup>2</sup>*Faculty of Biology, University of Gdansk, Gdańsk, Poland*

When we talk about fungal ecology, we think of disciplines that investigate the distinct ecological roles of fungi. Phytopathologists look for parasitic fungi. Research on mycorrhizae reveals the diversity of root-associated fungi. Plant physiologists unravel fungi hidden in plant tissues without obvious symptoms, lesions or symbiotic organs (e.g. root colonisation without mycorrhiza): these so-called endophytic fungi can influence plant physiology positively or negatively. Finally, dead organic matter reveals saprotrophic fungi. But what do fungal facts, especially in the metabarcoding area, tell us about this ?

An accumulating number of independent studies report fungi of 'known' ecological niche in bizarre, unexpected situations. Among these, many ectomycorrhizal fungi or ericoid/orchid mycorrhizal fungi are found as endophytes in roots of various, mostly herbaceous plants. Based on our recent research, we will provide direct and indirect evidence, both morphological and functional, for endophytism in diverse fungi, including *Tuber* spp., but also Sebaciniales or Russulales.

Finally, we propose an evolutionary interpretation of the ectomycorrhizal-endophytic niche: the evolution of the mycorrhizal habit through a pathway known as the 'waiting room' hypothesis (Selosse *et al.*, 2009). Root endophytism may act as a symbiotic 'waiting room', where biotrophic coexistence predisposes evolution towards a tighter mutualism with a more complex mycorrhizal morphogenesis. Some fungal taxa, now mycorrhizal, also retain the ancestral endophytic habit. The latter evolutionary pathway seems relevant for fungi mycorrhizal in ericoids and orchids, but also for some ectomycorrhizal taxa.

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## **Białowieża Forest: yesterday, today, tomorrow ...**

**Bogdan Jaroszewicz**

*Białowieża Geobotanical Station, Faculty of Biology, University of Warsaw, Białowieża, Poland*

Only some small isolated patches of forests with a high degree of naturalness survived till recently in Europe. Białowieża Forest is the last temperate forest of the European lowlands, where substantial fragments of well-preserved old growth have survived. With its continuous forest cover, documented by palynological studies, for close to 12,000 years, the forest is a very unique site in the continental and the world scale. Due to long lasting protection, started at the turn of 14<sup>th</sup> and 15<sup>th</sup> century, large portion of the forest is still composed of stands originating from the pre-silviculture period. The commercial logging started there as late as the beginning of the 20<sup>th</sup> century. In effect, ecosystems of Białowieża Forest are characterized by low level of anthropogenic conversion, the dominance of uneven age stands, great amount of deadwood, and high biodiversity. The patches of stands with high naturalness, still scattered throughout the commercial forest, guarantee that surrounding, culturally modified stands will recover soon after leaving them intact. As the stability of forest dynamics is scale-dependent, the expansion of a non-intervention approach is needed to increase the stability of the entire ecosystem and enhance the chances for its successful adaptation to changing environmental conditions. The non-intervention approach is of paramount importance, as natural forest once logged and artificially replanted, is gone forever or at least takes centuries to recover. Such forests are irreplaceable: they protect the diversity of specialized organisms and processes and provide valuable ecosystem services. They are priceless outdoor labs for ecological and evolutionary sciences, the benchmarks for conservation science and modern forestry.

## **Trees ancient and young: a veritable feast for wood decay fungi**

**Lynne Boddy**

*Cardiff University, UK*

A tree is never just a tree. It is always a tree plus a myriad of micro-organisms – especially fungi - operating together. The tree mycobiome comprises mycorrhizal fungi associated with roots; endophytes in all tree tissues – leaves, twigs, branches, trunks and roots; pathogens of leaves, above ground woody parts, and roots; and saprotrophs of all plant tissues. I will consider those fungi that decay wood, including saprotrophs, endophytes and pathogens. Wood is a very hostile environment, yet eventually every tree will be colonised by wood decay fungi. These fungi have adopted five main ways for colonising wood in standing trees, but these are not mutually exclusive: (1) heart rot; (2) wound colonizers; (3) active pathogenesis (i.e. killing living cells); (4) endophytes already present hidden within sapwood (latency); and (5) colonizers that come after other fungi (secondary colonizers). Research began in the mid 1800s, and centred around heart-rot, but more or less stopped 50 years ago, having made little progress. After early emphasis on heart rot, focus changed to decay following wounding in the 1960's and 70's, then in the 1980's to natural establishment in branches and trunks by fungi latently present in sapwood. Research has now come a full circle, with interest in heart-rot again. Pathogenesis has always been a priority, although people's perceptions of what are pathogens are often confused. I will talk about these complementary roles of fungi, and their different patterns of colonisation, in broadleaved trees. This will include our most recent findings on heart-rot in beech (*Fagus sylvatica*) and latent fungi, revealed by DNA analysis, and our approach to causing hollowing of younger trees to provide habitat for other organisms.



## FROM GENOME TO FUNCTION



OS - oral session

**Selective deconstruction of plant biomass: fungal enzymes active at the onset of solid state fermentation**

**Marie-Noëlle Rosso**<sup>1</sup>, Shingo Miyauchi<sup>1</sup>, Laurence Lesage-Meessen<sup>1,2</sup>, Anne Favel<sup>2,3</sup>, Elodie Drula<sup>1</sup>, Simeng Zhou<sup>3</sup>, Sacha Grisel<sup>1</sup>, Igor V. Grigoriev<sup>4</sup>, Robert Riley<sup>4</sup>, Isabelle Herpoël-Gimbert<sup>3</sup>, Sana Raouche<sup>3</sup>

<sup>1</sup>*INRA, Marseille, France*

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<sup>3</sup>*Aix-Marseille Université, Marseille, France*

<sup>4</sup>*Joint Genome Institute, University of California, Berkeley, USA*

White-rot fungi are the ultimate degraders of highly recalcitrant raw lignocellulosic substrates in forest ecosystems. White-rot fungi can degrade all plant cell wall polymers through the concerted secretion of complex sets of hydrolytic and oxidative enzymes. Among white-rot fungi, some species selectively degrade a larger proportion of lignin and hemicelluloses rather than cellulose, thereby keeping C6 saccharides preserved. These species have therefore been identified as interesting biological agents for the pretreatment of biomass dedicated to bioconversion.

We screened 63 white-rot fungal strains maintained at the CIRM-CF collection ([www6.inra.fr/cirm/Champignons-Filamenteux](http://www6.inra.fr/cirm/Champignons-Filamenteux)), for selective delignification of wheat straw under solid-state fermentation and identified *Polyporus brumalis* BRFM 985 as the best performing strain. The cultivation of the fungus for two weeks on wheat straw significantly improved the accessibility of cellulase-rich enzymatic cocktails to residual carbohydrates. In order to understand the lignin degrading capability of *P. brumalis*, we conducted an integrative multi-omics analysis by combining data from the genome, transcriptomes, and secretomes.

As a peculiar feature of *Polyporus brumalis*, we observed gene family expansion, up-regulation and secretion of an abundant set of Versatile Peroxidases and Manganese Peroxidases, compared with other Polyporales species. The orchestrated secretion of an abundant set of these delignifying enzymes and redox cycling enzymatic partners could contribute to the delignification capabilities of the fungus. Our findings highlight the diversity of wood decay mechanisms present in Polyporales and the potentiality of further exploring this taxonomic order for enzymatic functions of biotechnological interest.

OS - oral session

### **The evolution of mycorrhizal genomes and transcriptomes**

**Annegret Kohler**<sup>1</sup>, Eniko Kiss<sup>2</sup>, Emmanuelle Morin<sup>1</sup>, Shingo Miyauchi<sup>1</sup>, Laszlo G. Nagy<sup>2</sup>, Igor Grigoriev<sup>3</sup>, Francis Martin<sup>1</sup>

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<sup>2</sup>*Synthetic and Systems Biology Unit, Biological Research Center, Hungarian Academy of Sciences, Szeged, Hungary*

<sup>3</sup>*US Department of Energy, Joint Genome Institute (JGI), Walnut Creek, USA*

To elucidate the genetic basis and the evolution of the mycorrhizal lifestyle we took advantage of the large fungal genome sequencing programs ongoing at the Joint Genome Institute.

The comparison of 112 genomes from mycorrhizal fungi, wood decayers and litter decomposers revealed several independent lifestyle transitions from saprotrophism to mutualism. A general trend within ectomycorrhizal genomes, but not in ericoid and orchid mycorrhizal genomes, is the reduction of genes coding for plant cell wall degrading enzymes. However, ectomycorrhizal species seem to have retained diverse sets of enzymes depending on their ecological niches.

The availability of mycorrhizal genomes enables us as well to compare mycorrhizal transcriptomes and to identify symbiosis-related genes.

We could show that both conserved and clade-specific genes, as for example effector-like mycorrhiza-induced small secreted proteins (MiSSPs), are used by *Laccaria bicolor* to establish symbiosis with the roots of the host tree *Populus*. Using a phylostratigraphy approach we compared the « symbiosis-toolbox » of 15 mycorrhizal interactions. Again the age distribution of mycorrhiza-induced genes showed two peaks corresponding to ancestral genes made of conserved gene families and species-specific orphan genes.

About 45% of the mycorrhiza-induced genes predate the origin of mycorrhiza, suggesting gene co-option as a major mode of recruiting genes for symbiosis. Conserved mycorrhiza-induced genes showed little overlaps across species, suggesting that independently evolved mycorrhizal lineages have co-opted different genes. Interestingly, induced genes are coding for the same functions but without orthology, like small-secreted proteins, transporters or carbohydrate active enzymes.

OS - oral session

**Bioinformatics approaches to understanding regulation of secondary metabolism in non-model fungi**

**Ekaterina Shelest**

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Secondary metabolite (SM) genes are often clustered in fungal genomes, i.e., they are co-localized and co-regulated. About half of known SM gene clusters have an “own” embedded transcription regulator responsible for their induction. But in other cases, it is hard to find out how the expression of the cluster genes is coordinated, whether there is one single regulator, and which TF it could be. The situation becomes even more challenging in non-model fungi, especially in those that are not susceptible to genetic manipulations. In these cases, we have to rely on bioinformatics methods to reduce the experimental work and predict the most probable candidate regulators. One of the fruitful approaches is comparative genomics combined with promoter analysis and in this talk I will show how it can be applied to getting insights into mechanisms and evolution of SM regulation in fungi. The main assumption is that a TF may be embedded in the cluster or not, but the binding sites should be shared by all regulated genes promoters. If clusters (hence biosynthetic pathways) are well conserved, the upstream signalling pathways can be also conserved, and we can assume involvement of the same TF in the regulation in different species. Thus, applying comparative analysis, we come up not only with prediction of common motifs shared between the clusters but also with prediction of potential regulator or if the mode of regulation might differ between the species. For instance, with the help of promoter analysis we could show dissimilarity of pigment regulation in two basidiomycete fungi. Another example is the detection of common promoter motif indicating a general regulatory mechanism of atromentin synthetase genes for numerous basidiomycetes.

OS - oral session

**Fungal macrolactone toolkit: a phylogenomic atlas reveals usual suspects and unusual configurations in biosynthesis of ancient aromatic polyketides**

**Grzegorz Koczyk**<sup>1</sup>, Michał Kawałiło<sup>1</sup>, Zuzanna Dutkiewicz<sup>1</sup>, Monika Urbaniak<sup>1</sup>, Delfina Popiel<sup>2</sup>

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Fungal benzenediol lactones constitute a diverse class of secondary metabolites with many peculiar mechanistic and evolutionary features. Prior research has shown that biosynthesis of some compounds proceeds via bacterial rather than fungal cyclisation route and that the core biosynthetic polyketide synthase genes were likely a subject of frequent horizontal transfer among the filamentous fungi. Focusing on analysis of preexisting genomic resources and novel sequence data we show how macrolactone biosynthetic toolkit comes in both diverse and unexpected packages, with producers spread across the major orders of Ascomycetes.

On basis of fungal isolate screening, next generation sequencing, assembly and annotation of producer strains we demonstrate that biosynthesis of different macrolactone groups spans across divergent taxa, including evidence pointing to biosynthesis of dihydroxyphenylacetic acid lactones in members of the *Pochonia* genus. Through large scale phylogenomic reconstructions and reconciliations we have arrived at the annotation of macrolactone biosynthetic toolkit in the form of "phylogenomic atlas" a collected body of evidence which refines the timeline of key events contributing to lactone diversity, including tentative duplications and horizontal transfers as well as the recruitment of key accessory enzymes from different biosynthetic routes. As an illustrative example, we present evidence for recruitment and adaptation of key accessory FAD-dependent halogenase from biosynthesis of spirocyclic lactones such as geodin and griseofulvin, into the biosynthesis of halogenated resorcylic acid lactones.

Research supported by the National Science Centre (Poland) OPUS grant: 2016/21/B/NZ9/01875.

OS - oral session

### **Comparative genomics, evolution and adaptation of Antarctic cryptoendolithic Black Fungi**

Claudia Coleine<sup>1</sup>, Jason Stajich<sup>2</sup>, Sawyer Masonjones<sup>2</sup>, Laura Zucconi<sup>1</sup>, Silvano Onofri<sup>1</sup>, Laura Selbmann<sup>1</sup>

<sup>1</sup>University of Tuscia, Viterbo, Italy

<sup>2</sup>University of California, Riverside, USA

Black fungi are able to colonize a diverse array of inhospitable ecological habitats such as the ice-free areas of Continental Antarctica, including the McMurdo Dry Valleys, accounted as the coldest and most hyper-arid desert on Earth and the closest terrestrial Martian analogue. In these regions, these microorganisms find refuge inside rocks and they are commonly associated with cryptoendolithic communities, representing a border adaptation for life, before the extinction.

Studies on the genome evolution of these fungi may enable understanding of important genetic factors that govern their success in the extremes. In this work, we have assembled and annotated the genome sequences of Antarctic isolates (*Friedmanniomyces simplex*, *F. endolithicus*, *Hortaea thailandica*, *Rachicladosporium* sp., *R. antarcticum*, *Exophiala mesophila* and *Cryomyces minteri*) and compared them with other extremotolerant species. Genomes size ranged from 28.5 to 49 Mbp and predicted genes from 8,700 to 18,000. Gene function predictions were assigned by matches to the Pfam, MEROPS, CAZy, InterProScan and Swiss-Prot databases. Initial comparisons of gene and protein domain content among related species identified substantial gene novelty in the Antarctic strains, such as the pathways involved in the meristematic growth, DNA repair and cold-adaptation.

Moreover, since duplicate gene burden is a recurrent character in stress-adapted microbes, we also focused on Whole Genome Duplication analysis: in all cases the genomes and predicted proteomes contained more than 90% of identifiable fungal Universal Single-Copy Ortholog genes and in most of all cases, more than 50% were duplicated.

OS - oral session

### **Transposable elements in fungal genomes**

**Anna Muszewska**<sup>1</sup>, Kamil Steczkiewicz<sup>2</sup>, Marta Stepniewska-Dziubinska<sup>1</sup>, Krzysztof Ginalski<sup>2</sup>

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Transposon (TE) insertion can detach co-selected traits, alter gene transcription, lead to chromosomal rearrangements and introduce insertional mutations. In the last decade, remarkable examples of the functional impact of TEs on the host's fate have been described. Specifically, the effects of transposon proximity have been studied mainly for model species. We executed several comparative genomics projects exploring TEs in hundreds of fungal genomes. TEs were predicted using both de novo and homology-based approaches and screened against known protein domains related to TEs for additional confirmation.

Predominantly, we found that fungi associated with a plant host tend to accumulate DNA transposons and retrotransposons with long terminal repeats (LTRs). In addition, TE composition can be a species-specific trait and thus determination of TE suite can be useful for cryptic species delimitation. Some types of transposons are present only in old lineages (Mucoromycotina, DIRS), while others can be found solely in Dikarya genomes (Ngaro).

We could confirm previously developed models of genome evolution assuming a simultaneous increase in the number of copies of various mobile elements. We have also observed that the presence of complementary genome protection systems, i.e. an RNAi-based system or Repeat-Induced Point mutations correlate with the decline of active TEs in favour of fragmented, inactive copies. Moreover, remnants of TEs massively overlap with regions annotated as genes, while younger, potentially active TEs cluster together away from genic regions. Observed non-random distribution of potentially active transposons might be a sign of selection against insertion of TEs in gene proximity or target site preference among some types of TEs.



OS - oral session

**Cytogenomic analyses reveal nuclear content variation along the life cycles of the Pucciniales fungi and enable the distinction of heterokaryotic nuclei**

**Rita Carvalho**<sup>1</sup>, Helena Azinheira<sup>1,2</sup>, Marta Monteiro<sup>3</sup>, João Loureiro<sup>4</sup>, Leonor Morais-Cecílio<sup>1</sup>, Pedro Talhinhos<sup>1,2</sup>

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Fungal life cycles are characterised by predominant haploid stages, while in most cases diploid nuclei occur only in a single cell, following karyogamy between haploid nuclei and immediately followed by meiosis. Rust fungi are also reported as obeying to this general rule, with the basidium as the single diploid cell. However, recently flow cytometric studies showed the presence of 1C, 2C and a low proportion of 4C nuclei in different stages of the urediniosporic cycle of several rust fungi. While cryptosexuality phenomena and the formation of uredinoid teliospores have been suggested to explain such results, endopolyploidy or other parasexuality phenomena cannot be ruled out.

Moreover, the identification of a rust species (*Coleosporium tussilaginis* f. sp. *senecionis-silvatici*) containing equal quantities of slightly different (in size) nuclei suggests that these could represent each of the nuclei in the dikaryotic cell. If so, then this ‘twin peaks’ phenomenon enables the isolation of sister nuclei, representing an unprecedented tool for studying sexuality/mating types and other aspects of heterothallism, in fact, RNAseq performed on RNA extracted from FACS-separated nuclei populations enabled us to assign different pheromone receptor alleles to each of the nuclei populations.

This study intends to clarify the ‘twin peaks’ phenomenon and the nuclear cycle of rusts by transcriptomic and cytogenomics techniques approaches as FISH in sorted nuclei in which our results strongly suggest the occurrence of diploid nuclei.



PS - poster session

### **Identification of Antibacterial Agents from East African Basidiomycota**

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The Basidiomycota comprises the mushroom-forming fungi and various other organisms that represent a considerable part of the global mycodiversity. Recent molecular ecology studies have revealed huge diversity of Basidiomycota in different habitats, including soil, plants, and invertebrates.<sup>1</sup> Most of these fungi and especially the majority of the species from tropical areas have neither been cultured nor studied for potential beneficial traits such as the production of antibiotics or other useful secondary metabolites.<sup>2</sup> During the course of searching for new anti-infective and other biologically active secondary metabolites from Kenyan Basidiomycota, 13 previously undescribed metabolites, (6R,7S,10R)-7,10-epoxy-7,11-dimethyldodec-1-ene-6,11-diol (1) and 12 sesquiterpenes named elgonenes A–L(2–13), and the known compound p-coumaric acid (14) were isolated from a basidiomycete collected in Mount Elgon Natural Reserve.<sup>3</sup> The producing organism represents a new species of the genus *Sanghuangporus*, which is one of the segregates of the important traditional Asian medicinal mushrooms that were formerly known as the “*Inonotus linteus*” complex. Twelve previously undescribed metabolites named skeletocutins A-L (1-5, 7-13) together with the known tyromycin A (6) were isolated from a *Skeletocutis* sp. (Polyporales), also originating from Mount Elgon. The isolation, structure elucidation and biological activities of these secondary metabolites will be presented.



PS - poster session

**Diversity and pathogenicity of the *Alternaria* species complex involved in apple leaf blotch and fruit spots in France**

**Kévin Fontaine**<sup>1</sup>, Céline Fourrier-Jeandel<sup>1</sup>, Manuela Dagba<sup>2</sup>, Anne-Laure Boutigny<sup>1</sup>, Valérie Caffier<sup>3</sup>, Jason Shiller<sup>3</sup>, Bruno Le Cam<sup>3</sup>, Renaud Ioos<sup>1</sup>, Jaime Aguayo<sup>1</sup>

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Apple leaf blotch and fruits spots caused by *Alternaria* species are common diseases in apple production regions. Several *Alternaria* species or complex species including *A. arborescens*, *A. longipes*, *A. gaisen* and *A. alternata* seem responsible of fruit losses and decrease tree vigor.

Serious defoliations linked to *A. alternata* f. sp *mali* have been observed in the past in Japan and US. This fungus classified as quarantine pathogen (A1 list of EPPO, 2000/29/CE directive) carries a gene cluster involved in the biosynthesis of a host-specific toxin: AM-toxin. However, several cases of *Alternaria* leaf blotch have been reported recently which suggests a possible emergence of this pathogen in Europe.

The goal of this study is to achieve the taxonomic assignation of >200 *Alternaria* isolates responsible for symptoms observed in France, by a multilocus sequence typing approach. A specific detection of *Alternaria* f. sp *mali* was performed by targeting three genes involved in AM-toxin biosynthesis. Pathogenicity assays of 30 strains collected worldwide was conducted on detached leaves to compare their virulence on Golden and Gala varieties.

Preliminary prospections of orchards showed that the fruit spots disease seems to represent a minor problem in France. On the other hand, leaf spots have been annually observed, with different levels of severity, on 10 different apple varieties. Collected isolates corresponded mostly to *A. arborescens* and to the *A. alternata* complex. Neither *Alternaria* f. sp *mali* nor *Alternaria gaisen* were isolated from French orchards.

Finally, pathogenicity assays suggest that all strains may produce symptoms with a great variability within strains, regardless if they carry or not the gene cluster involved in the production of the AM-toxin.

PS - poster session

**Waste wood bioconversion under oxygen depletion by *Phlebia radiata* - Towards sustainable bioethanol**

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*University of Helsinki, Helsinki, Finland*

Wood decomposing Basidiomycota Polyporales are a familiar sight on tree trunks. Wood decomposition itself is taking place beneath the bark and even inside waterlogged pieces of wood. We hypothesized that the wood-decay fungal hyphae face partial oxygen depletion. Therefore, we investigated how oxygen-depletion affects the fungal decomposition of wood leading to intake and bioconversion of the released sugars into ethanol. The study is part of our research aiming at sustainable second-generation bioethanol production.

Our work started by a screening of white rot Polyporales fungi for their capability of simultaneous saccharification and fermentation (SSF). On waste lignocelluloses, *Phlebia radiata* 79 outperformed other candidates in ethanol bioconversion (Mattila et al. 2017, 2018). We proceeded by studying the transcriptomics and regulation of CAZYme and core metabolic genes of the fungus by RNA-sequencing.

At first, we learned that expression of CAZYme encoding genes is affected by atmospheric changes. Hypoxia is seemingly an equally important regulator as the lignocellulose substrate. The multiple cellulose, hemicellulose, pectin as well as lignin targeted genes responded specifically to hypoxia. In some cases, CAZY genes and the respective intracellular catabolic pathway shared similar expression pattern pointing to co-regulation of extra- and intracellular fungal metabolism.

Next, the promoter regions of intracellular core metabolic and CAZYme encoding genes were studied in silico. We noticed that response to hypoxia in *P. radiata* is similar to the Basidiomycota human and animal pathogenic *Cryptococcus* species than e.g. Ascomycota species. In addition, regulation of acetyl-CoA and glycerol metabolisms are essential in the adaptation to hypoxic conditions.

PS - poster session

**Special membrane domains of *Schizophyllum commune*, similar to lipid rafts?**

**Jessica Pötschner**, Erika Kothe  
*Friedrich Schiller University, Jena, Germany*

The basidiomycete *Schizophyllum commune* belongs to the white rot fungi and is relevant for wood degradation worldwide. Its high competitive ability is based on the recognition of other fungi and bacteria, the production of specific extracellular metabolites and a strategy of fast growth. Therefore, fast transport processes within the hyphae and an efficient cytoskeleton are essential.

Here we investigated special membrane domains of the model organism *S. commune*. Fluorescent dyes like filipin have been used to visualize sterol-rich domains in the membrane of this fungus. A continuous flow of secretion vesicles from the hyphal cell body to the growing hyphal tip provides the delivery of proteins and lipids to the tip and is essential for cell wall and cell membrane extension at the tip. Lipid rafts are involved in the transport of these essential proteins. Furthermore, detection and identification of lipid raft localized signal proteins were performed. Investigations about localization of special membrane domains in *S. commune* showed similarities to lipid rafts, whereas insights into formation and protein association revealed differences to lipid rafts of e.g. humans or ascomycetes.

PS - poster session

**Insights in hyphal development of *Schizophyllum commune***

**Elke-Martina Jung**<sup>1</sup>, Marjatta Raudaskoski<sup>2</sup>, Erika Kothe<sup>1</sup>

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The white rot basidiomycete *S. commune* is relevant for wood degradation worldwide. As early colonizer of tree wounds and after forest fire, the fungus has also phytopathogenic importance. Its high competitive ability is based on the recognition of environmental signals from other bacteria and fungi as well as the production of specific extracellular metabolites like laccases and a strategy of fast growth.

The fungal cytoskeleton, composed of a complex network of microtubules and actin structures, has a major impact on transport of vesicles as well as endo- and exocytosis processes. Visualization of the actin cytoskeleton in actively growing hyphae was performed with Lifeact-GFP. Thereby cortical actin patches were visualized at cell tips and clamps and as well as in subapical cells, preceded septation. The actin cytoskeleton in living hyphae during septum development shows close association with nuclear division. Clamp cell formation, typical for many basidiomycetes including the model fungus *S. commune*, indicated an aggregation of actin filaments to ring structures at the future site of nuclear division. Additionally, GFP-labeling of histone H2B enables visualization of nuclear movement and mitosis events in monokaryotic and dikaryotic cells. After mating events, fast nuclear exchange in anastomoses and hyphal cells were observed.

PS - poster session

**Unique wood-decay strategies and fruiting body development in the Schizophyllaceae**

**Neha Sahu**

*MTA-SZBK Biological Research Centre, Szeged, Hungary*

Agaricomycetes are fruiting body forming fungi that produce some of the most efficient enzyme systems to degrade woody plant materials. Despite decades-long interest in the ecological and functional diversity of wood-decay types and in fruiting body development, the evolution of the gene repertoires of both traits are incompletely known. Here, we sequenced and analyzed the genome of *Auriculariopsis ampla*, a close relative of the model species *Schizophyllum commune*. Comparative analyses of wood-decay genes in these and other 29 Agaricomycetes species revealed that the gene family composition of *A. ampla* and *S. commune* are transitional between that of white rot species and less efficient wood-degraders (brown rot, ectomycorrhizal). Rich repertoires of suberinase and tannase genes were found in both species, with tannases generally restricted to species that preferentially colonize bark-covered wood. Analyses of fruiting body transcriptomes in both *A. ampla* and *S. commune* highlighted a high rate of divergence of developmental gene expression. Several genes with conserved developmental expression were found, nevertheless, including 9 new transcription factors as well as small secreted proteins, some of which may serve as fruiting body-specific effector molecules. Taken together, the genome sequence and developmental transcriptome of *Auriculariopsis ampla* has highlighted novel aspects of wood-decay diversity and of fruiting body development in mushroom-forming fungi.

PS - poster session

**Evolutionary history of a common fungal endophyte of Norway Spruce**

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Fungal endophytes live asymptotically in plants. While the myriad of diverse fungi found within host plants certainly play important ecological roles to the host, their biological attributes remain a riddle to unveil. In the northern hemisphere, *Lophodermium piceae* is probably the most common and abundant foliar endophytic fungus of Norway Spruce. To understand the dispersal and the evolutionary history of this widespread fungus, we investigated the population genomics and mycelial morphology of *L. piceae* across the entire native distribution range of its host, *Picea abies*. We sampled 20 trees in each of the 31 natural forests in Europe. From spruce needles, we purified and isolated ca. 600 *L. piceae* isolates and confirmed their identification by sequencing the whole Internal Transcribed Spacer (ITS) region. We phenotyped all the isolates using a digital image-based approach. Using restriction site associated DNA sequencing data (RAD seq) generated from 475 isolates, we investigated the spatial population structure and phylogeography of this endophyte. Preliminary results suggest that *L. piceae* exhibits high levels of both genetic and phenotypic variation but divergent *L. piceae* lineages. These results provide a fundamental foundation for future studies on local adaptation, colonisation history and the ecological role of fungal endophyte-host interactions.

PS - poster session

### **Revision of A1 family of aspartic proteinases in basal fungi**

**Małgorzata Zielińska**, Anna Muszewska  
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Aspartyl proteases A01 (AP) show great differences in abundance between fungi displaying distinct ecological strategies. Members of this family can be found in many organisms ranging from mammals to retroviruses and plants. It is hypothesized that early functions of APs may have been related to the external digestion of food proteins by saprophytic organisms. In recent years, new fungal homologs bearing unique sequence motifs were identified. In consequence, a fine-grained subfamily classification of A1 family could have been established. This work focuses on the attempt to deepen and systematize the knowledge of ancient fungal A1 peptidases and to trace their evolution within the kingdom Fungi.

To achieve this goal we identified representatives belonging to A1 family in proteomes of 45 basal fungi and 10 model Dikarya species. In the next steps, we discarded inactive homologs and performed a phylogenetic analysis of over 730 AP sequences. The results were compared to the contemporary classification of A1 family in eukaryotes.

Our results significantly expand and thoroughly revise existing A1 classification. Most of the identified clades are corresponding to those previously described. Only two of previously identified six subfamilies seem to have emerged after the divergence of Dikarya and three were likely lost in this lineage. Basal taxa are present in most clades and form three specific clades. One of them groups a huge expansion of *Batrachochytrium* sequences what is of particular interest because aspartyl proteases are involved in *Batrachochytrium* infections. Several other A01 expansions are present among analysed fungi and may play also a role in nutrient acquisition and pathogenesis.





PS - poster session

**Effects of abiotic stresses on ketoisovalerate reductase (KIVR) gene expression and cyclodepsipeptides production by fungi from Hypocreales order**

**Monika Urbaniak**<sup>1</sup>, Grzegorz Koczyk<sup>2</sup>, Agnieszka Waśkiewicz<sup>3</sup>, Zuzanna Dutkiewicz<sup>2</sup>, Łukasz Stępień<sup>1</sup>

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Fungi belonging to Hypocreales order are among the most prolific producers of secondary metabolites, including the non-ribosomally produced depsipeptide mycotoxins beauvericin (BEA), enniatin (ENN) and their analogues. Core cyclodepsipeptides are synthesized by modular megasynthases (NRPS, non-ribosomal peptide synthases). However, multiple accessory genes can function at different stages of biosynthesis both for product modification and for procurement of substrates. In case of depsipeptides, the ketoisovalerate reductase (KIVR) provides hydroxyisovaleric acid, the availability of which functions as a bottleneck of the pathway.

In our study we investigated the producers of different cyclic depsipeptides (enniatis, beauvericin) among entomopathogenic (*Beauveria*, *Metarhizium*, *Cordyceps*, *Isaria*) and phytopathogenic (*Fusarium*) representatives of ecologically diverse Hypocreales order. We analysed whether abiotic stresses may affect KIVR gene expression and subsequent production of different cyclodepsipeptides.

Six fungal strains were cultivated on modified liquid medium. Salinity stress was induced using 0.7 M solution of sodium chloride, whereas osmotic stress was induced using 30% solution of polyethylene glycol 6000. RT-PCR analysis was performed for KIVR gene and normalized against tub2 gene expression in 3 biological replicates. For cyclodepsipeptides quantification LC/MS/MS system (Waters, Milford, PA, USA) was used.

Correlations between expression and toxin profile are discussed in the context of interspecies differences, shedding more light on the shared trait of depsipeptide production by divergent Cordycipitaceae.

Research supported by Polish National Science Centre PRELUDIUM 13 grant: NCN 2017/25/N/NZ9/02525.



PS - poster session

**Patterns of cytogenomic variability across fungi and the occurrence of very large genomes in the Pucciniales**

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The completion of genome sequencing for some rust fungi has contributed to suggest a link between biotrophic specialisation and genome size expansion. The measurement of genome sizes for 60 rust fungi using Flow Cytometry has revealed some of the largest genomes among fungi, with nine rust species with haploid genomes between 300 and 2489 Mbp, with an average of ca. 380 Mbp. Genome size information is available for over 1800 fungal species, either arising from Flow Cytometry, genome sequencing, or other methods, yielding a global average of 60 Mbp. Departing from our genome size measurements of Pucciniales fungi, in this work we analyse genome size variability across representatives of the entire fungal phylogeny and attempt to relate such variations with relevant biological and genomic traits (life style, nutrient use, composition in transposable elements, etc.). The analysis of genome size variation can unveil clues suggesting polyploidisation events or transposable elements activity of evolutionary/adaptive relevance. Such traits can be associated with reproduction strategies (sexual, asexual, parasexual and/or rare sexual) and substrate utilization (saprobes, mutualists, obligate/facultative pathogens, biotrophs/necrotrophs and combinations of those). In average, fungi that interact with plants have larger genomes than those that interact with animals or other fungi (88, 27 and 29 Mbp, respectively), while those feeding on non-living substrates present intermediate values (53 Mbp).



PS - poster session

**Genetic diversity of fungal community in soil amendment with mineral fertilizers enriched with microorganisms**

Agata Gryta, Magdalena Frać, Karolina Oszust, Mateusz Maćcik, Giorgia Pertile  
*Institute of Agrophysics, Polish Academy of Sciences, Lublin, Poland*

Fungi are important component of soil microbiome. Fungal communities are involved in circulation of elements and improving plant growth. The activity of fungi as well as bacteria and archaea is responsible for biological soil status. Genetic diversity of soil fungal communities depends on physical and chemical properties of soil with different fertilization. After application of fertilizers to soil, the fungal community can change throughout the soil profile.

The aim of the study was to evaluate the changes in community of soil fungi after application of mineral fertilizer enriched with microorganisms. In the field experiment, the optimal dose of fertilizer was used without microbial enrichment, the optimal dose containing microorganisms and the reduced dose by 40% microbiologically enriched.

Molecular approaches such as t-RFLP is technique that has been used to search microbial communities to determine the diversity of phylogenetic. In performed research, DNA was directly extracted from 0.5 g of soil using a bead-beating method (FastDNA SPIN Kit for soil, Bio 101 Inc., USA). PCR based on amplification with specific fluorescent labelled primers (ITS1 f \*6-FAM CTTGGTCATTTAGAGGAAGTAA; ITS4 r TCCTCCGCTTATTGATATGC (Gardes and Bruns, 1993)) was applied to the study on soil fungi. The purified products were then digested with endonucleases Hae III. Subsequently, digested products were separated and their size were detected on automated sequencing capillary electrophoresis.

The obtained t-RFLP peaks profile indicated shifts in the fungal community structure of tested treatment. The DNA patterns of amended soil were characterized by few high peak and a lot of small peaks. It shows that soil fungal community is very diverse and consist with a few major group and many minor group.

## TAXONOMY AND SYSTEMATICS

OS - oral session

**Recent developments in the (infrageneric) classification of *Aspergillus* and *Penicillium***

**Jos Houbraken**<sup>1</sup>, Robert A. Samson<sup>1</sup>, Jens C. Frisvad<sup>2</sup>

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The family of Aspergillaceae harbors various economically important genera, such as *Aspergillus*, *Penicillium* and *Monascus*. Recently, new insights in the taxonomy of those genera have led to numerous new species and name changes of existing species. Current taxonomic studies have shown that species delimitation in *Penicillium* is rather clear cut with no intergrading strains and only a few hybrids have been reported. Of course individual characters may be overlapping, such as the size, form and ornamentation of conidia or growth and sporulation as related to temperature, but a large number of characters are non-overlapping. This is especially true concerning small molecule extrolites characters. Species are thus stable for many years (punctuated equilibria), and may then speciate under certain circumstances. However, there is one problem with the use of clear non-overlapping characters: these characters are not necessarily present in all isolates of a species or all species in a series, section, subgenus or genus. This phenomenon may be caused by horizontal transfer of genes or gene clusters or of epigenetical factors or mutation based loss of certain gene clusters. Some of the more interesting categories are those between the genus and the species: subgenera, sections, subsections, series and subseries. Do these levels have nomenclatural status, are they predictive and should they be used formally in taxonomy? We present examples of these categories and discuss whether these categories between genus and species are useful in the taxonomy of *Penicillium*, *Aspergillus* and *Monascus*. Examples in e.g. sections *Roquefortorum*, *Sclerotiora* and *Brevicompecta* will be given.

OS - oral session

**Anybody seen root galls on graminoid plants? First steps towards understanding the evolution of inconspicuous *Entorrhiza* and its relatives**

**Kai Riess**<sup>1</sup>, Max E. Schoen<sup>2</sup>, Max E. Schoen<sup>2</sup>, Matthias Lutz<sup>3</sup>, Roger G. Shivas<sup>4</sup>, Marcin Piątek<sup>5</sup>, Sigisfredo Garnica<sup>6</sup>

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*Entorrhiza* is a fungal genus that causes galls on roots of Cyperaceae and Juncaceae. The few known species are nearly globally distributed but rarely studied due to their hidden lifestyle that causes no aboveground visible effects on host plants. Although *Entorrhiza* was identified 130 years ago, crucial questions about the origin, phylogenetic relationships or biology remained unanswered.

The establishment of a new phylum related to the Dikarya, Entorrhizomycota, prompted a renewed exploration of this enigmatic fungal group. The existing molecular and ultrastructure data suggest two likely hypotheses: (i) *Entorrhiza* appeared as a sister group to the rest of the Dikarya, or (ii) *Entorrhiza* forms a clade with the Basidiomycota. As a next step, *Talbotiomyces* was identified as the nearest relative to *Entorrhiza*. However, both taxa represent a deep dichotomy within the Entorrhizomycota. Interestingly, the host range was thereby broadened from monocotyledons (*Entorrhiza*) to eudicotyledons (*Talbotiomyces*). Recent topology-based and distance-based co-phylogenetic analyses revealed a deep phylogenetic split corresponding to species infecting Cyperaceae (*Entorrhiza* s. str.) or Juncaceae (*Juncorrhiza* gen. nov.). This observation was also congruent with the spore morphology. Relaxed molecular clock analyses suggested that the Entorrhizomycota originated around the Neoproterozoic-Paleozoic. Within *Entorrhiza* and *Juncorrhiza*, the major radiation took place during Oligocene-Miocene.

Due to the arcane lifestyle of *Entorrhiza* and its relatives, most of the diversity is still to be discovered. Therefore, more root examinations of potential host plants are needed. Finally, the fungal biology needs to be better understood and to clarify their ecological significance.

OS - oral session

***Chalara* revival – how to treat chalara-like fungi?**

**Ondřej Koukol**

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Species of the anamorphic genus *Chalara* are characterized by phialidic conidiogenous cell that has broad venter and long tubular collarete and produces hyaline, cylindrical (non)septate conidia. The distinct chalara-like morphology evolved multiple times and chalara-like anamorphs are currently placed in more than ten orders within Pezizomycotina. Though currently classified as members of Microascales (IndexFungorum), majority of *Chalara* species belong to the order Helotiales. Although known teleomorph connection and molecular data allow proper phylogenetic placement of these species and new distinct lineages with chalara-like morphology are accommodated in separate genera, the delimitation of *Chalara* s. str. remains unknown. This lack of interest is connected with predominantly saprotrophic strategy of majority of species, their high morphological similarity resulting in numerous misidentified records and rather rare occurrence. During previous decade, new anamorph-teleomorph connections were recorded and molecular data obtained for common and also rare species allowing their phylogenetic placement. Based on these recent findings together with literature review on ultrastructural data, the delimitation of *Chalara* s. str. and evaluation of morphological trends among particular chalara-like lineages will be discussed.

OS - oral session

**Polythetic taxonomy of Iranian Hypoxylaceae and Xylariaceae of the Xylariales**

Mohammad Javad Pourmoghaddam<sup>1</sup>, **Christopher Lambert**<sup>2</sup>, Frank Surup<sup>2</sup>, Kathrin Wittstein<sup>2</sup>, Marc Stadler<sup>2</sup>,  
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During a survey of Iranian Hypoxylaceae and Xylariaceae, new species for the Iran as well as species new to science have been described via a polythetic taxonomical approach, covering chemotaxonomy of fruiting bodies via high performance liquid chromatography with diode array coupled mass spectrometry (HPLC-DAD/MS), morphological descriptions via light- and scanning electron microscopy (SEM) and multilocus phylogenetic inference with four different gene loci; the internal transcribed spacer region (ITS), the large subunit (LSU) of the nuclear rDNA, beta-tubulin (TUB2) and the second largest subunit of the RNA polymerase II (RPB2). A Comparison of the chemotaxonomical data with our internal database gave rise to several unprecedented metabolites which have been subsequently isolated via a combination of preparative HPLC and thin-layer chromatography (TLC). Structural assignment followed by nuclear magnetic resonance (NMR) spectroscopy lead to 10 newly described compounds from environmental sources. Phylogenetic inference showed three hotspots of genetical diversity in respect to reference data, further fortifying the segregation of species from their corresponding complexes inside *Hypoxylon fuscum* and *H. rubiginosum*. Furthermore, Iranian *Dematophora* with synnemmatized *Geniculosporium*-like anamorph and *Rosellinia* without synnemata have been characterised by multilocus genealogy and chemically by investigation of the secondary metabolite profiles of representative strains, confirming the segregation of *Dematophora* and *Rosellinia* as shown by Wittstein et al. 2019.



OS - oral session

**Phylogenetic revision of the basal Dacrymycetes (Basidiomycota): Cerinomycetaceae family**

**Anton Savchenko**

*University of Tartu, Tartu, Estonia*

Dacrymycetes (Basidiomycota) is phylogenetically and morphologically well-defined class, sister to the Agaricomycetes. It has diverged from the other extant fungi approximately 350-380 million years ago, and supposedly included the first brown rot wood-decomposers in the kingdom. The class is relatively small and currently comprises ~110 species in three families. Most members of the class have gelatinous basidiocarps of different yellow tints due to a presence of carotenoids. The traditional generic delimitation was established on a basis of basidiocarp types and has remained unchanged since the mid of 20<sup>th</sup> century. In modern phylogeny-based works this approach has been shown to be outdated since all of the major genera appeared to be polyphyletic. We present here a revision that clarifies generic concepts in the basal Dacrymycetes lineages, and reestablishes family division for the class. We put a special focus on Cerinomycetaceae family, the largest basal group and the second-largest family after Dacrymycetaceae. In the result we restrict it to a single genus *Cerinomyces* with 32 species (22 of them undescribed). We also reflect on the most ancient lineages in the class, namely Unilacrymaceae and related newly discovered taxa, and compare them to Cerinomycetaceae.

We show that micromorphology is far more important as a genus-defining character than basidiocarp type. For example, we transfer several species with gelatinous basidiocarps to traditionally arid-corticoid *Cerinomyces*, and exclude from the genus several corticioid species that clustered to Dacrymycetaceae. The hidden diversity uncovered in the study suggest that further works in basal Dacrymycetes still promise a large number of taxonomic findings in the future.

OS - oral session

**Patterns of speciation in thallus-forming Laboulbeniomyces**

**Danny Haelewaters**

*Purdue University, West Lafayette, USA*

Our understanding of fungal diversity is far from complete. Species descriptions generally focus on morphological features, but this approach may underestimate true diversity. Using the morphological species concept, *Hesperomyces virescens* (Ascomycota, Laboulbeniales) is a single species with global distribution and wide host range. Since its description 120 years ago, this fungal parasite has been reported from 30 ladybird host species on all continents except Antarctica. These host usage patterns suggest that *H. virescens* could be made up of many different species, each adapted to individual host species. Using sequence data from three loci, we found distinct clades within *Hesperomyces virescens*, each corresponding to isolates from a single host species. We employed sequence-based species delimitation methods, which support that these lineages represent separate species, driven by adaptation to different ladybird hosts. In contrast, using the same approach, we found that *Rickia wasmannii*, which parasitizes 10 phylogenetically segregated species of *Myrmica* ants, is a single phylogenetic species. As we are exploring more examples of Laboulbeniales fungi, we can start linking speciation patterns to presence/absence of traits. One candidate trait that may have an influence in host-dependent speciation in Laboulbeniales is the presence of a haustorium. Haustoria are rhizoidal structures that can be simple or branched and penetrate the host's integument to provide additional holdfast and to increase surface area, presumably for nutrient uptake.

OS - oral session

**New taxa of Antarctic cryptoendolithic black fungi by multi locus phylogeny**

**Laura Selbmann**, Federica Mostardi, Claudia Coleine, Laura Zucconi, Silvano Onofri  
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Black Fungi, also known as Rock-inhabiting black fungi (RIF) or Microcolonial Fungi (MCF) are specialized in colonizing almost every extreme environment on Earth, including extremely dry habitats as cold and hot deserts where they exploit endolithic niches as main refuge for life. In the Antarctic ice-free areas, as for the McMurdo Dry Valleys, they take part of the cryptoendolithic communities, representing a border life-style for microbes living in constant weak balance between life and extinction. Black fungi (RIF) in these communities are highly resistant and adapted organisms and there they live in spatial association with similarly extremotolerant organisms. The geographic and genetic isolation and the unique environmental pressure led to the evolution of extremely adapted and specialized genotypes and these ecosystems represent a rich reservoir of still unknown fungal biodiversity.

In the present study, we describe a relevant number of new RIF genera and species, defined by a multilocus phylogeny in rock samples collected along an altitudinal transect from 1,000 to 3,300 m asl in Victoria Land (Continental Antarctica) during the XXXI Italian Antarctic Campaign.

All new taxa were included in Dothideomycetes; in particular, 2 new genera and 4 new species were sitting in the Teratosphaeriaceae, 3 new species were discerned in the genus *Friedmanniomyces* (Teratosphaeriaceae), 1 new species in the genus *Rachicladosporium* (Cladosporiaceae) and 1 in the genus *Cryomyces* (incertae sedis).

PS - poster session

**Two new African siblings of *Pulveroboletus ravenelii* (Boletaceae)**

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Several books on West African mycology mention *Pulveroboletus* aff. *ravenelii*, a species of bright yellow color and quite common in the Guineo-Sudanese and Zambezian forests.

The study aims to clarify the taxonomy of species associated with *Pulveroboletus ravenelii* based on morphological and genetic characters.

We used a combination of anatomorphologic characters, as well as phylogenetic analyzes of DNA sequences from 41 African *Pulveroboletus* specimens and compared to those from North American and Asian species to describe the two new species. Amplification and sequencing of the *atp6*, *tef1* and *rpb2* genes were performed using primer pairs ATP6-1M40F and ATP6-2M, EF1-983F and EF1-2218R, and bRPB2-6F and bRPB2-7.1R.

Phylogenetic analysis has shown that African specimens form a sister subclade of Asian and American taxa. Two new African species, *Pulveroboletus africanus* sp. nov. and *Pulveroboletus sokponianus* sp. nov. are described and illustrated. The African collections represent two separate species, *P. africanus* sp. nov. and *P. sokponianus* sp. nov. both macroscopically similar to *P. ravenelii*.

Both new species are endemic to tropical Africa. Finally, phylogenetic analysis shows that all specimens of *Pulveroboletus* studied, including the new species, form a strongly supported monophyletic clade (BS = 100%).

It is interesting to note that specimens of the new African species form a distinct subclade (BS = 100%), separated from the sibling subclade formed by American and Asian species.

PS - poster session

### **Six new species in *Aspergillus* section *Nidulantes***

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*Aspergillus* section *Nidulantes* encompasses both homothallic and anamorphic (probably heterothallic) species mostly occurring in soil, plant material or indoor environment. Some of them are clinically relevant or produce mycotoxins. The objective of this study was to reevaluate the species boundaries within several clades of the section *Nidulantes* using the available strains from culture collections as well as newly isolated strains. We assembled five datasets containing the new isolates and their closest relatives and performed phylogenetic and phenotypic analysis. We tested the hypotheses if the newly isolated or re-examined strains constitute separate species (splitting approach) or should be treated as part of broadly defined species (lumping approach). Four DNA sequence loci were amplified, i.e. ITS and LSU rDNA and partial sequences of the *benA*, *CaM* and *RPB2* genes. The latter three loci were used for the phylogenetic analysis and served as the input for single-locus (GMYC, bGMYC, PTP and bPTP) and multi-locus (STACEY and BP&P) species delimitation methods. The phenotypic analysis comprised macro- and micromorphology (including SEM) and comparison of maximum growth temperature. The phylogenetic analysis supported the splitting hypothesis in all cases and based on the combined approach, we propose six new species, three of which are homothallic and three anamorphic. Four from the six newly described species were isolated from the indoor environment (Jamaica, Trinidad & Tobago, USA), one originated from soil (Australia), and one from the Kangaroo rat (USA).

PS - poster session

**New record of *Hyphoderma etruriae* (Fungi, Basidiomycota) in Portugal and its combination in *Lawrynomycetes***

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*Hyphoderma etruriae* Bernicchia is a very rare corticioid species, described originally from Italy and more recently reported from Macedonia, always growing on decayed wood of *Juniperus* spp. Here we provide a further recent record from Lisbon, encountered on a stump of *Cupressus lusitanica*. *H. etruriae* differs from other species of *Hyphoderma* s.l. on account of a grandinioid hymenophore, clampless hyphae, hyphoid capitate cystidia and incrustated hyphal endings.

Based on its microscopic characteristics, several authors have suggested that *H. etruriae* could be related to *Hyphoderma capitatum* J. Erikss. & Å. Strid and *Hyphoderma orphanellum* (Bourdot & Galzin) Donk. and our molecular analyses confirmed that these three species belong to the Hymenochaetales and are thus not closely related to *Hyphoderma* s. str. species (Larsson 2007; Bernicchia & Gorjón 2010). The monospecific new genus *Lawrynomycetes* was already proposed by Karasinski (2013), to accommodate *H. capitatum*. According to our multigene phylogenetic analyses (18S, 5.8S, 28S, RPB2), *H. etruriae* is in a well supported clade with *H. capitatum*, both species sharing clampless hyphae, capitate cystidia and an association with conifers. On the contrary, our results do not support a close relationship between *H. orphanellum* and the *H. capitatum*-*H. etruriae* clade. Based on our results, the new combination *Lawrynomycetes etruriae* (Bernicchia) Melo, Salcedo & Olariaga is proposed.

PS - poster session

**Using variable rate models to study the evolution of ectomycorrhizal fungi through time**

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Fungi form one of the most diverse kingdoms in terms of both functions and species. They can get their nutrients from living organisms (biotrophic) or dead matter (saprotrophic). The most common biotrophic lifestyle is mycorrhiza which is a symbiotic root association that allows the uptake of nutrients by plants. Ectomycorrhiza involves the largest number of mycorrhizal fungi and is essential for many stand forming trees. However, the evolutionary history of ECM is still unresolved. It is clear that ECM evolved several times in both plants and fungi, but there are two conflicting views about the evolution of ECM in fungi suggesting (1) relatively few transitions to ECM followed reversals to saprotrophy or (2) >78 independent origins and no reversals of ECM. In this study we investigate the patterns of evolution of ECM and check if the ECM clades evolved independently or if there have been any reversals to saprotrophic lifestyle. We assembled a densely sampled dataset of 2,174 fungal taxa covering the three subphyla that include ECM fungi: Pezizomycotina, Agaricomycotina, and Mucoromycotina. We created a time-calibrated phylogeny using MCMCTree and R8s for a concatenated dataset of 6 marker gene sequences (SSU, 5.8S, LSU, rpb1, rpb2, tef) where all taxa must have LSU sequence. Previous studies addressing this question have either just done informal parsimony like analyses, or have assumed a constant rate of evolution through time. As ECM plants have not been available until the Late Triassic or Early Jurassic, and the area dominated by ECM plants has varied during the evolutionary history of fungi it seem reasonable that the rate has varied through time too. We therefore investigated the evolution of ECM fungi using model-based methods with variable rates through time.

PS - poster session

**Disentangling the identity of the genus *Biatorellina***

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The monospecific genus *Biatorellina* is currently considered a taxonomic synonym of *Trybliopsis* but has an obscure and complicated history. During the type revision of the genus *Tympanis*, an isotype of *Biatorellina buchsii* was fortuitously found and reviewed. Initially our revision led to the hypothesis that *B. buchsii* could be conspecific with *Tympanis confusa*. A bibliographic and morphological revision, together with a biometric study, was done using the isotype of *B. buchsii* and specimens of *Tympanis confusa* to verify the identity of the genus *Biatorellina*. Our results show an overlap in the morphology, biometry, distribution and ecology of *B. buchsii* (= *T. buchsii*) and *T. confusa*. The identity and the placement of *Biatorellina* is resolved and *B. buchsii* is proposed as a synonym of *T. confusa*.



PS - poster session

### What do we know about *Grovesiella*?

Luis Quijada<sup>1</sup>, Eugene Popov<sup>2</sup>, Adam Polhorský<sup>3</sup>, Joey Tanney<sup>4</sup>, **Jiří Kout**<sup>5</sup>, Peter R. Johnston<sup>6</sup>, Donald Pfister<sup>1</sup>

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The two accepted species of the genus *Grovesiella* are pathogens that grow on conifers in temperate North America and Europe. *Scleroderris abieticola*, the basionym of the type species is from North America. Morelet combined in the genus *Grovesiella* in 1969. After that three further species were combined or described in the genus. Currently, of these species only *G. grantii* A. Funk remains in the genus. It is differentiated from *G. abieticola* by its smaller apothecia, asci and ascospores. In 1993 Sieber and Kowalski pointed out differences in the ascospores biometry among collections of *G. abieticola* from Europe and North America. Three groups were differentiated in their biometric analysis, two for North America and one from Europe, each group growing on a different species of *Abies*. Also, they explained that the collections from Europe do not produce cankers while the North American populations do. In 2015, Baral & Quijada placed *Grovesiella* in the family Tympanidaceae based mainly on morphological resemblances, but there is no multi-gene phylogeny that has used sequences to verify this relationship. Finally, two years ago Chastagner et al. (2017) showed that the ITS sequences of *G. abieticola* from Europe and North America are only 83% similar. Taking all of this in consideration, we for first time use sequences, type revisions and morphology-biometric studies to answer the following questions: Is *Grovesiella* properly placed in Tympanidaceae? Do these taxa represent a single genus? How many species can we differentiate in the genus?

PS - poster session

**First worldwide report of *Colletotrichum tamarilloi* causing anthracnose in strawberry**

**Lincoln Vicente Araújo do Bizerra**<sup>1</sup>, Fernanda Lara dos Santos<sup>1</sup>, Danilo Batista Pinho<sup>1</sup>, Ailton Reis<sup>2</sup>,  
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Several important strawberry pathogens belong in the genus *Colletotrichum*, due to the genus high aggressiveness and capacity to infect different plant organs and to cause several diseases, including anthracnose, crown rot and flower blight. *Colletotrichum* is responsible for significant economic losses in the strawberry growing regions worldwide. Since different species of *Colletotrichum* are associated with strawberry diseases in different plant parts, precise species identification is essential for the recommendation of efficient control strategies and, eventually, to the reduction of pesticide residue in the commercial produce. Therefore, the project aims to determine the species of *Colletotrichum* occurring in strawberry fields in Brazil. Fruits, flowers and strawberry crowns showing typical symptoms of *Colletotrichum* infection were collected in commercial fields in the administrative regions of Brazlândia and Recanto das Emas in Distrito Federal; in the municipalities of Atibaia in São Paulo; São Francisco de Paula in Rio Grande do Sul; Castelo and Domingos Martins, in Espírito Santo; and Goianópolis and Padre Bernardo, in Goiás. In total, 50 isolates were obtained and had genomic DNA extracted. The amplification and sequencing of the GAPDH gene region of all the isolates was performed in order to carry out the preliminary identification. The ITS and  $\beta$ -tubulin gene regions of nine representative isolates were amplified and sequenced for more detailed identification and phylogenetic analysis. While most samples yielded *C. nymphaea*, the species *C. tamarilloi* was identified and associated with strawberry symptoms in one region (Atibaia), representing the first report of *C. tamarilloi* associated with strawberry worldwide.

PS - poster session

***Phytophthora abietivora*, a new species isolated from diseased Christmas trees in Connecticut, USA**

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A number of fir species, (*Abies* spp.), are produced as Christmas trees around the world. In particular, the Fraser fir, *Abies fraseri* (Pursh) Poir., is popular as it yields high-quality Christmas trees in temperate North America and Europe. A *Phytophthora* sp. causing root rot on Fraser fir was isolated from a Christmas tree farm in Connecticut, USA, and found to be new to science according to morphological and molecular phylogenetic analysis using multi-locus DNA sequences from ITS, Cox1,  $\beta$ -Tub, Nadh1, and Hsp90 loci. Thus, it was described and illustrated as *Phytophthora abietivora*. An informative Koch's postulate test revealed that *P. abietivora* was the pathogen causing root rot of Fraser fir.

PS - poster session

**Diversity and phylogeny of the compactoid *Russula*'s**

**Ruben De Lange**, Annemieke Verbeken  
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Within the genus *Russula*, one of the major ectomycorrhizal fungal genera in all ecosystems worldwide, the compactoid *Russula*'s (*R. subg. Compactae* s.l.) are a large and atypical group, mainly characterised by the presence of lamellulae which are absent in the rest of the genus. The fruitbodies are rather big, firm and compact with a short and thick stipe and a white, yellowish or brown cap. The context is often browning or blackening, sometimes reddening. The subgenus was recently split into four subgenera: *R. subg. Compactae* s.s., *R. subg. Malodora*, *R. subg. Brevipes* and *R. subg. Archae*. The objective of this project is to delimit species within these subgenera in Europe and place them in a worldwide context. We aim to unravel the species complexes and describe new species through phylogenetic and morphological analysis. As it is thought that ecology might play an important role in speciation, we also focus on mycorrhizal host associations. An analysis was done on the existing data and a phylogenetic tree was created. Based on this tree we assume the existence of at least 38 undescribed European species within *R. subg. Compactae* s.l. in our current sequence dataset. This dataset contains sequences mostly from Europe, some from Brazil, Martinique, Thailand, Panama, North-America and Africa.

PS - poster session

**Barcoding approach as an effective tool for revealing *Astraeus* species in the Republic of North Macedonia**

**Katerina Rusevska**<sup>1</sup>, Mitko Karadelev<sup>1</sup>, María P. Martín<sup>2</sup>

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Until Phosri et al 2013, *Astraeus hygrometricus* was considered widely distributed around the world; however the authors found that *A. hygrometricus* in its strictest sense is, for example, not found in USA. Between the specimens analyzed these authors described a new species to Europe (*A. telleriae*) and two new species to USA (*A. morganii*, *A. smithii*) With the aim of clarifying the number of *Astraeus* species in the Republic of North Macedonia, and to verify some previous collections, molecular analyses were carried out on 64 samples from different localities and habitats in the country; including various type of forest associations, such as oak, hornbeam, beech, chestnut, juniper, pine, as well as meadows, pastures, etc.. Sixty new ITS sequences (Fungus barcode; Schoch et al. 2012) were generated, and compared with homologous sequences located in public databases. The different molecular analyses performed (e.g. parsimony, Bayesian) grouped the Macedonian specimens in different clades. According to the barcoding sequences, four *Astraeus* species are found in the country: *Astraeus hygrometricus*, *A. pteridis* and *A. telleriae*; as well as an *Astraeus* new species (Crous et al., in press).

This research received support from the SYNTHESYS Project ES-TAF-788, which is financed by the European Community Research Infrastructure Action under the FP7 ‘Capacities’ Program. The authors thank to the Biology Students’ Research Society and Macedonian Ecological Society for arranging many collecting trips resulting in valuable collections.

PS - poster session

**Systematic revision of the genera *Holwaya* and *Patinella* (Ascomycota) based on phylogenetic traits**

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The genus *Holwaya* was until recently considered monospecific with many taxa treated as synonyms of *H. mucida*, except for *H. pusilla* that was transferred to the genus *Claussenomyces*, and *H. salicis* to *Durandiella*. *Holwaya* was previously considered a member of the family Bulgariaceae based on morphological and DNA data (Wang et al. 2006, Lumbsch & Huhndorf 2010), but it was removed from this family by Crous et al. (2014) who regarded Bulgariaceae as a synonym of Phacidiaceae. In the latest classifications of Ascomycota, Baral (in Jaklitsch et al. 2016), followed by Wijayawardene et al. (2017), placed the genus in Tympanidaceae. Yet, the revision of Tympanidaceae (Quijada et al. in prep.), and the results of the 15 genes phylogeny of Leotiomycetes made by Johnston et al. (in press), showed that the genus is not related to *Tympanis*, but formed a sister clade to Thelebolales in Leotiomycetes as incertae sedis (Johnston et al. 2019). Here we provide the morphological and phylogenetic evidences that link the genera *Holwaya* and *Patinella*. The aim of this work is to improve the classification of Leotiomycetes by placing two genera with currently uncertain position into a new family with the aid of polyphasic taxonomy.

PS - poster session

**Species diagnostics of European oak powdery mildews**

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Powdery mildews of the *Erysiphe* sect. *Microsphaera* are the major foliar parasites of oaks. Three phylogenetic species (*Erysiphe alphitoides*, *E. hypophylla* and *E. quercicola*) have been identified based on molecular analysis on leaves of European oaks. These three species are closely related but they are considered to be cryptic and they use to be identified by DNA barcoding currently. Low fungal biomass, hyperparasites and other common fungal contaminants and inability to cultivate powdery mildews in vitro make DNA extractions and sequencing difficult. In this study we focused on verification of diagnostic morphological characters used in the literature and on identification of other possible characters suitable for the species recognising. Morphological characters observed on herbarium specimens and new field collections identified based on ITS sequences were statistically evaluated and we present significant morphological differences suitable for identification of three European oak *Erysiphe* species. Mixed infections of single oak leaves by both *E. alphitoides* and *E. quercicola* were found.

The study was supported by the Slovak Research and Development Agency under the grant no. APVV-15-0210.

PS - poster session

**New interesting records of the genus *Entoloma* from southern and insular Greece**

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*Entoloma* is a highly species-rich genus of the order Agaricales (Fungi, Basidiomycota) and accurate determination of species by delimiting intraspecific from interspecific morphological variation is often very difficult. Pertinent research in the western Mediterranean has led to the description of several species new to science; however, this genus remains poorly surveyed and inadequately studied in the eastern part of the Basin and particularly in typical Mediterranean habitats. The results presented here, are the outcome of a multi-disciplined approach, involving examination of both fresh material and exsiccatae, ecological features, nrDNA ITS sequencing and phylogenetic analysis. Three thermophilous species, namely *Entoloma olivaceohebes*, *E. sanvitalense* and *E. vindobonense* were found to be associated with Mediterranean *Pinus halepensis* forests in the Corinthos area (Peloponnese), and constitute new additions for the Greek mycobiota. Moreover, *E. cistophilum*, *E. llimonae* and *E. philocistus* associated with *Cistus creticus* from Paros Island (South Aegean Sea) are also presented, being the only confirmed records of the species in Greece to date. In addition, some of the studied specimens seem to represent so far undescribed species. For all aforementioned taxa, the phylogeny, biogeography and taxonomy are discussed and critical morphoanatomical features of pertinent specimens are illustrated.



PS - poster session

**Recent progress in species delimitation in *Subulicystidium* (Trechisporales, Basidiomycota)**

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Diversity of corticioid fungi (resupinate Basidiomycota), especially outside the northern temperate climatic zone, remains poorly explored. For many decades, the delimitation of species in *Subulicystidium* (Hydnodontaceae, Trechisporales) was a challenge. Presence of numerous transitional forms as to basidiospore size and shape hindered species delimitation, and almost no data on molecular diversity has been available.

We aimed at clarifying the species boundaries and distribution patterns in *Subulicystidium* and providing key for species identification.

Between 2015 and 2019, we examined ca. 200 *Subulicystidium* specimens mostly from Paleo- and Neotropics but some also from temperate and boreal Eurasia. We recorded size and shape of basidiospores and cystidia in a systematic way, and applied DNA barcoding and phylogenetic Bayesian and Maximum likelihood approaches to assess the variation in nrDNA (ITS and 28S regions) in the whole genus. We described 11 new species and thus doubled the number of known species in the genus *Subulicystidium*.

The main deliverables from analyses of ITS and 28S nrDNA in *Subulicystidium* are:

- (i) There is no universal barcoding gap applicable for the whole genus;
- (ii) At the level of individual species, a barcode gap is observable in *S. fusisporum*, *S. parvisporum*, *S. robustius* and *S. tedersooi*;
- (iii) Species *S. longisporum*, *S. obtusisporum*, *S. brachysorum* and *S. meridense* are highly polyphyletic;
- (iv) Species *S. perlongisporum* is monophyletic.

We found species with transoceanic distribution (e.g. *S. oberwinklerii* and *S. perlongisporum*) but also species known from the single isolated area so far (e.g. *S. parvisporum* on Réunion Island). The true distribution patterns of *Subulicystidium* species are yet to be elucidated.

PS - poster session

### **Morphology standards for description of *Russula* species**

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Morphological descriptions of *Russula* species (Russulales, Agaricomycotina) show distinct inconsistencies among authors and regions. Globally, *Russula* descriptions (i) are incomplete or differ in selection of structures and morphological characters; (ii) are inconsistent in terminology; (iii) have insufficient statistical support; (iv) use different techniques and chemical reagents; and (v) have low quality of illustrations. As a part of our initiative named “Quest of globally comprehensible *Russula* language” we organised the “*Russula* microscopy workshop” at February, 23<sup>rd</sup> – 28<sup>th</sup> 2018 in Banská Bystrica, Slovakia. Participants from different countries dealing with *Russula* collections from different continents discussed minimal requirements and standards for morphological description of *Russula* species. As a result, we proposed unified *Russula* terminology and description templates. These standards were applied for description of 23 new species and three re-described *Russula* species from South-East Asia, Australia, Europe, North and Central America. *Russula seperina* is presented here as an example of a description using the proposed morphology standards.

The organisation of the “*Russula* microscopy workshop” and this study is supported by the Slovak Research and Development Agency, grant no. APVV-15-0210.

PS - poster session

**The expanded diversity of Trichomeriaceae: *Knufia* and *Trichomerium***

**Magdalena Owczarek-Kościelniak**, Paweł Czachura, Marcin Piątek

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The family Trichomeriaceae described by Chomnunti and K.D. Hyde in 2012 includes 11 genera – *Arthrocladium*, *Bradomyces*, *Brycekendrickomyces*, *Hyalocladosporiella*, *Knufia*, *Lithohypha*, *Metulocladosporiella*, *Neophaeococcomyces*, *Neostrelitziana*, *Strelitziana* and *Trichomerium* – which exhibit diverse habitats and life strategies. Among them, *Knufia* and *Trichomerium* are the largest, the most divergent and widely distributed genera containing 13 (all sequenced) and 30 species (8 species sequenced), respectively. They are important rock inhabitants and plant colonizers causing sooty mould diseases. Recently, we have isolated numerous new strains of *Knufia* and *Trichomerium* from stones and sooty mould communities in Poland. In order to assign the strains to particular species, their rDNA ITS and LSU sequences together with related sequences (including environmental sequences) from GenBank were analysed and used for phylogenetic reconstruction. Species diversity based on barcoding regions is discussed, including species new for science recorded in materials from Poland.

The study was funded, in part, by the National Science Centre, Poland, under project 2017/27/B/NZ9/02902. It also received financial support, in part, from the W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, as part of the program for „Young Researchers”.

PS - poster session

**The genus *Rachicladosporium*: introducing two new species from sooty mould communities**

**Marcin Piątek**, Paweł Czachura, Magdalena Owczarek-Kościelniak  
*Department of Mycology, W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, Poland*

The species composition of sooty mould communities covering leaves of deciduous trees in temperate climate regions is weakly known. It is believed that in these regions the sooty moulds are predominantly composed of *Aureobasidium* and *Cladosporium* species. The species diversity in these communities is however much larger and, except common species, includes also rare species. The species of the genus *Rachicladosporium* were previously not reported as inhabitants of sooty mould communities. This genus was described by Crous, U. Braun and C.F. Hill in 2007. Originally it contained only one species, *Rachicladosporium luculiae*, isolated from leaf spots on plant *Luculia* sp. in New Zealand. Additional species were described later from different substrates, habitats and environments, including plant leaves and needles, twig, black mould on baobab trees and rocks. Currently the genus contain 13 species. Here, we introduce two new species that were isolated from sooty mould patches on leaves of woody plants in Poland. Both species were delimited based on morphological and molecular phylogenetic analyses. Our results expand evolutionary diversity and ecological preferences of the genus *Rachicladosporium*.

The study was funded by the National Science Centre, Poland, under the project 2017/27/B/NZ9/02902.

PS - poster session

***Ganoderma lucidum* (Curtis) P. Karst. sensu stricto: host tree and habitat range in Finland**

**Pyry Veteli**, Marta Cortina-Escribano, Henri Vanhanen, Riikka Linnakoski  
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Several temperate taxa in the genus *Ganoderma* P. Karst are often reported to occur on dead and declining coniferous trees. Historically, in many cases the assumed strict host preference has been considered as diagnostic taxonomic importance. The type species *G. lucidum* was originally described from *Coryllus avellana* in UK. However, general knowledge in Nordic countries is that *G. lucidum* is commonly found on *Picea abies*. In Finland, at the northernmost limits of the species' distribution, *G. lucidum* has been reported as a sporadically occurring species, typically in moist, fertile sites, associated with broadleaved trees. To our knowledge, a thorough inquiry into the ecology and the possibility of intraspecies diversity of *G. lucidum* in Nordic countries is yet to be undertaken.

Here we present first insights from our field inventories on several sites with abundant *G. lucidum*, accompanied by results from somatic compatibility of isolates, as well as mating tests between single spore isolates from conifer and hardwood-inhabiting strains. Multi-locus phylogenetic analyses and morphological comparisons from a wide range of strains collected from conifer and hardwood substrates in Finland is constructed to place the material into European and global context. On study sites with more than 10 colonized stumps per hectare, 56-60% of the stumps represented *P. abies* under fibre analysis. The single spore isolates from conifer and hardwood-inhabiting strains were found to be compatible.

The results discussed in the presentation serve both as valuable observations of ecological plasticity of wood decaying fungi, as well as informing the development processes of new fungal-based biotechnological applications such as commercial agroforestry practices.

PS - poster session

**Living at the extremes: : novel lineages in the Myriangiales**

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*Department of Mycology, W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, Poland*

The order Myriangiales consists of 13–16 genera, of which representatives of six genera (*Anhelia*, *Elsinoë*, *Endosporium*, *Myriangium*, *Phaeosclera*, *Sarcinomyces*) have already been sequenced and included in molecular phylogenetic analyses. Representatives of this weakly known yet important order are mostly connected with plants, mainly as pathogens which cause numerous plant diseases. In our recent studies we have isolated some novel strains of the Myriangiales from extreme habitats: resin exudates of *Pinus sylvestris* and epiphytic sooty mould communities. All samples were collected in Central Europe. The generic and specific affinities of these strains are evaluated by integrative taxonomy including morphology and DNA sequence data. This study extends the knowledge of the Myriangiales by detecting novel lineages in this order and showing interesting extreme habitats for its representatives.

The study was funded, in part, by the National Science Centre, Poland, under project 2017/27/B/NZ9/02902. It also received financial support, in part, from the W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, as part of the program for „Young Researchers”.

PS - poster session

**Cryptic diversity within the smut fungal species *Anthracoidea sempervirentis***

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The smut fungal genus *Anthracoidea* (Ustilaginomycotina, Ustilaginales) contains more than 100 species that parasitize hosts predominantly in the sedge genus *Carex*. *Anthracoidea* species are mainly found in the boreal zones of the Northern Hemisphere and many species have an arctic-alpine distribution. Host specificity for many of the species in this genus is considered to be quite broad and a host spectrum of over ten host species is common, although such a pattern is uncommon in other plant parasites. Host specificity and species delimitations for many *Anthracoidea* species, however, are so far primarily assessed based on morphological data, such as spore shape, spore ornamentation and spore size. In this study we try to understand host specificity and species delimitation in *Anthracoidea sempervirentis*, a prevalent species occurring on different host species in the *Carex* Section *Aulocystis* using molecular data. Some of the host species are prevalent members of the plant community in the Alps and/or the Balkan Peninsular, where they can have overlapping geographic ranges and may occur in close spatial proximity to each other. The aim of this phylogenetic study based on the rDNA large subunit is therefore to understand whether species delimitation in *Anthracoidea sempervirentis* correlates with host identity or whether similar host ecologies have led to parasite species with broad host ranges.

PS - poster session

**The genus *Ravenelia* in South Africa – Diversity, Phylogeny and population structures**

**Malte Ebinghaus**<sup>1</sup>, Martin Kemler<sup>1</sup>, Andreas Brachmann<sup>2</sup>, Michael Wingfield<sup>3</sup>, Wolfgang Maier<sup>4</sup>, Domink Begerow<sup>1</sup>

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The rust fungi are the most diverse group of obligate plant parasitic fungi and commonly infect the prevailing number of land plant lineages throughout major climatic regimes. The genus *Ravenelia*, however, appears to be confined to members of the legume family with tropical and sub-tropical distributions. With more than 250 species, it is the third largest genus of rusts and can easily be recognized due to its notably complex and distinguished teliospores. Here, we will present recent research on the species diversity and phylogeny of South African representatives of *Ravenelia*. Furthermore, the results of an additional population genomic study on the gall-rust *R. macowaniana* that was based on a ddRAD-approach will be presented. Our research on the diversity and phylogeny of *Ravenelia* led to an increase of species numbers from 19 recognised species to now 32 *Ravenelias* in South Africa. The phylogenetic reconstructions show also evidence for rust lineages specifically associated to monophyletic host groups but furthermore indicate a polyphyletic origin of *Ravenelia*. In our population genomic study we implemented samples of *R. macowaniana* on its two hosts *Vachellia karroo* and *V. natalitia* collected from 22 sites throughout South Africa. These results show a differentiation of this species into at least 3 distinct and geographically separated populations and no clear pattern of host specificity could be found.



PS - poster session

**A post-harvest disease of vanilla caused by two new *Moniliella* species**

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Moniliellomycetes is an enigmatic group of yeasts within Ustilaginomycota. No sexual state has been reported for these fungi so far. Currently, the class comprises one genus with 18 species. These species have been described from mainly two habitats: flower nectar and fermenting substrates such as meat and cheese. Several, *Moniliella* species, especially *M. megachiliensis* are commercially used in the food industry for the production of erythritol. Here we report the finding and phylogenetic placement of two presumably new *Moniliella* species from vanilla pods. The species were observed on high-value vanilla pods that had been vacuum-sealed and were ready for resell. The occurrence of the yeasts rendered the pods not marketable. This is the first time that representatives of this small basidiomycetous class of fungi are reported to be involved in post-harvest spoilage of plant material.

PS - poster session

**Evolution of ascospore morphology in bryophilous Pezizales**

**Lukáš Janošik**

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Shape of the actively ejected ascospores of Ascomycetes is under a strong selective pressure on minimization of their drag. However, only little is known about the other factors responsible for the great diversity of ascospore morphology. Bryophilous Pezizales represent a species rich monophyletic group of several genera associated with bryophytes, which members exhibit highly diverse ascospore morphology, ecology and represent unique model group for the research of ascospore evolution.

Combination of the ascospore ejection experiments, detailed morphometric analysis and molecular data were used to elucidate which other factors affect their dispersal and could play important role in the ascospore evolution.

Number of nuclei influenced size of the ascospores in several species with variable number of ascospores in asci. Presence of the ornamentation and organization of the ascospores in asci were also found to have strong effect on the spore cluster formation, what could affect their ability to colonize new localities. Interestingly the species with smooth ascospores are usually connected to the stems or leaves of their host, whereas the species with ornamented ascospores were usually restricted only to the rhizoids.

PS - poster session

**Species diversity of myxomycetes as a challenge for modern molecular technics**

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Myxomycetes (also known as a slime molds) are known from middle ages based from description of *Lycogala epidendrum* in 1654. This group of organism include about 1000 species known to now. Due to the complexity of the ecological/morphological characteristics, their taxonomic classification provoke many problems. The development of molecular techniques is an important contribution to the study of the taxonomy of myxomycetes and allows to obtain very specific data from DNA analysis. Based on small subunit ribosomal DNA sequences we have made an attempt to species identification one of slime mold observed in Białowieża Forest. Comparative DNA analysis confirms similarity to *Fuligo septica* species and the closest variant of genetic similarity pointed *Fuligo septica* var. *flava*. However, belonging to this clade seems unlikely due to the value of the genetic distance, which is equal of 17,7 %, with 7 indels included (based on total length of common sequence 488 bp). What's more, almost identical (different only by one nucleotide) DNA sequence, as obtain in our study, was already deposited in GenBank by Hoppe (2018). Although, the inability of species identification caused to leave this record with unclassified status. The use of molecular research usually works well to identify species or subspecies, however complexity and variability of myxomycetes definitely requires further study in a very wide spectrum of different techniques to receive answers to appearing questions about taxonomic status.

PS - poster session

**Multilocus phylogeny of Laboulbeniomyces (Ascomycota) with special emphasis on Pyxidiophorales**

**Michał Gorczak**, Kamil Kisło, Marta Wrzosek

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Laboulbeniomyces, a class of ascomycetous fungi, has been poorly represented in major molecular phylogenetic studies resolving evolutionary relationships in fungi. Similarly relationships between its three orders are unresolved. Two of them – Laboulbeniales and the recently described Herpomycetales – are obligate biotrophic ectoparasites of arthropods. Less is known about the free-living, mycelial order Pyxidiophorales. The main objectives of this study were to resolve phylogenetic relationships among lineages of Laboulbeniomyces and to test possible evolutionary scenarios of transition from polymorphic hyphal organism to obligate ectoparasite.

Newly generated marker sequences were obtained from cultures of Pyxidiophorales and unculturable specimens of other Laboulbeniomyces. Phylogenetic trees were generated using maximum likelihood and Bayesian approaches based on three ribosomal DNA markers (SSU, ITS, LSU) and TEF1 $\alpha$ .

The addition of TEF1 $\alpha$  sequences and better sampling of Pyxidiophorales significantly increased support for major branches in Laboulbeniomyces phylogenetic reconstructions. Our analyses provide strong support for the class Laboulbeniomyces and the orders Herpomycetales and Pyxidiophorales.

## FUNGI IN BIOTECHNOLOGY

OS - oral session

**Symbiotic fungi as an efficient tool in biotechnology**

**Katarzyna Turnau**

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A wide range of microbes can be used in cost-efficient and sustainable techniques in agriculture, phytoremediation, agromining and plant protection. Selection of the most efficient endophytic and mycorrhizal fungi assisted by symbiotic bacteria can be used to speed-up plant growth, to limit the use of fertilizers and pesticides and to defend plants from pathogens. Above mentioned interactions can be additionally supported by animals that have been found to be an important factor serving as propagule vectors and sources of key nutrients.

Bacterial and fungal endophytes were isolated from plants colonizing post-flotation wastes rich in heavy metals, agricultural and agromining areas, and extremely harsh sites. None of these plants were devoid of microbiota. Many of them were strongly mycorrhizal but this type of symbiosis can be hampered due to wrong use of agrotechnology or presence of soil toxicity. The endophytic fungi in roots are generally more common than mycorrhizae. In addition, they often colonize the shoots, although, their potential in phytoremediation is still mostly neglected. Nonmycorrhizal plants depend only on endophytic microbes. These are very important in optimization of Ni agromining in Europe. Fungal symbiotic endophytes are widely considered for their potential in biotechnology, thus understanding the mechanisms of endophyte dependent growth acceleration in plants seems essential. In most cases the communication between microbes and plants, between microbes themselves (also including interactions between fungi and bacteria) are of crucial importance in developing the most effective technologies supporting diverse plant communities and forming “infrastructure” between diverse plants.

OS - oral session

**Inositol signaling in *Schizophyllum commune***

**Erika Kothe**, Reyna Murry, Elke-Martina Jung, Katrin Krause

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Phosphatidyl inositol phosphate is involved in eukaryote cellular signaling including the mammalian CREB pathway and lithium therapy for mental disorders. Using the basidiomycete *Schizophyllum commune*, first evidence was obtained showing involvement of inositol signaling in sexual development. Cross-talk to Ras signaling could be established via inositol monophosphatase (IMPase), down-regulated in strains with constitutive Ras1 signaling. Lithium repression of IMPase enzyme activity induced a massive increase in the levels of highly phosphorylated inositol species that could not be established using other model organisms including *Cryptococcus* or *Saccharomyces*, or mammalian cells, although that had been postulated from the inositol depletion theory. Proteome analysis indicated a shift of fungal metabolism and cellular signaling upon lithium application.

The results of inositol signaling in cell morphology and physiology in the filamentous basidiomycete was investigated, and links to cytoskeleton, endocytosis, or pheromone signaling were observed. Since cytoskeleton functions were shown to be involved in clamp formation, development and the fast nuclear migration involved in mating interactions was addressed. A very fast transport of nuclei is required to establish the dikaryon, which may involve a new cytological feature singular to basidiomycetes. This might explain the unprecedented finding of split dynein heavy chain genes in this fungal clade.



OS - oral session

**Development of a biotechnology tool for tree stress mitigation under urban context**

**Miguel A. Ramos**<sup>1</sup>, Cindy Serafim<sup>1</sup>, Tugce Yilmaz<sup>1</sup>, Kang Yu<sup>2</sup>, Maarten Van Geel<sup>2</sup>, Ben Somers<sup>2</sup>, Thierry Ameglio<sup>3</sup>, Olivier Honnay<sup>2</sup>, Paula M.L. Castro<sup>1</sup>

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Trees are key elements in mitigating the common environmental problems in urban areas, provisioning crucial ecosystem services such as air quality improvement, decrease of water runoff and microclimate mitigation. Ectomycorrhizal Fungi (ECM) may play an important role in urban tree management, improving tree vigor, and thus the extent of ecosystem services delivered by urban trees under stress. An in-depth exploitation of the potential of dedicated EcM inocula as the active factor in management of trees in urban ecosystems through an analysis of tree physiology, growth response remains to be done.

The major aims of this work were to assess the growth performance of *Tilia tomentosa* inoculated with ECM species and study plant response to water stress. An 9 months in-vivo experiment was established with seedlings exposed to different substrate pH (Acid and Alkaline), 5 inoculation treatments and different water stress conditions (no stress and stress).

Inoculation of *T. tomentosa* seedlings with selected inocula significantly promoted height growth, diameter, foliar dry weight of seedlings in acid substrate. SPAD levels were measured at different time points in a stress dynamic experiment (before stress, water stress, recover). Under the stress period a protection/mitigation effect was observed from ECM to the plants in acid and alkaline substrate. Plant that were exposed to water stress have normalized their SPAD values under recovery and at the end stage.

These results represent an important contribution for development and application of ECM inocula in urban context.





OS - oral session

**Screening of strains from wood decay species for advancement of mycelium-based materials technology**

**Carolina Girometta**<sup>1</sup>, Stefano Babbini<sup>2</sup>, Rebecca Michela Baiguera<sup>1</sup>, Diego Savio Branciforti<sup>1</sup>, Marco Cartabia<sup>1,2</sup>, Daniele Dondi<sup>1</sup>, Anna Maria Picco<sup>1</sup>, Elena Savino<sup>1</sup>

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Wood decay fungi (WDF) mostly include species in Agaricomycetes. Besides the chemical degradation, WDF hyphae mechanically force the lignocellulosic matrix by high turgor pressure. Several WDF species develop macroscopic, annual or perennial basidiomes, whose consistency is notably tenacious, flexible or rigid, rubber-like or wood-like. Analogous features are displayed by mycelia too, as appreciable in pure culture. If allowed to colonize a 3d-substrate (e.g. saw dust), the fungus gradually replaces the substrate itself and produces a strongly cemented, composite material. If the fungal biomass is completely separable from the substrate (e.g. liquid medium), the final material is composed by mycelia only.

Mycelia and its biocomposites were reported in literature for promising physico-mechanical and thermodynamic properties with concern to: low density; thermal and acoustic insulation; fire safety; compressive, tensile and flexural strength; elastic deformation. In- and extra-wall panels, floor tiles, design objects and packaging boxes are examples of mycelium-based products.

Whatever is the parameter regarded, the pivotal variable is always the fungal strain. Aims of our research are: isolation of mycelia in pure culture; molecular check of morphological identification; screening based on mycelial mat morphology, selectivity towards substrate, growth rate, cultivation parameters.

ITS region analysis resulted indispensable to discriminate cryptic species. Preliminary screening on synthetic media was followed by growth tests on specialized substrates based on the species preference in wild. The selectivity towards different substrate components (basically, cellulose vs lignin) was investigated by coupling thermogravimetric analysis (TGA) and infrared spectroscopy (FTIR).

OS - oral session

**Taming the monster or how to control fungal morphology in the submerged cultures**

**Marcin Bizukojć**

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Submerged cultures are the common method of filamentous fungi cultivation to produce primary and secondary metabolites, and enzymes. These cultivations are run in various types of bioreactors, including stirred tank bioreactor, air-lift bioreactors, slurry (bubble) columns or shake flasks. Nevertheless, all these fungal processes are not easy to run as their main problem is to assure the appropriate amount of oxygen for their growth and metabolite formation. This problem is mainly caused by the varying rheological properties of the fungal broths. In consequence mixing and mass transfer processes are aggravated and it is all for the development of complex fungal morphology in the submerged cultures. Filamentous fungi (and filamentous microbes like actinobacteria) tend to form various morphological forms from dispersed growth to the agglomerates of various size and structure. Furthermore, the morphological form developed has, in most cases, the direct association with the productivity of metabolites. All these make the need to control fungal morphology and few morphological engineering techniques were proposed within the last decade. To them belong microparticles-enhanced cultivation (MPEC), changing of broth surface tension, viscosity and osmolality. Genetic modifications of microorganisms were also proposed. Last but not least, if a fungus is co-cultivated in the two-species cultures, its first response to the presence of another microorganism is the change of its morphology.

Three aims of this review work are: (1) to show the importance of fungal morphology in the submerged cultures with regard to the overall run of the process, (2) to have a deep insight into the morphological development of various species and (3) to present the morphological engineering techniques to influence on the morphological development of the individual species.

Summing up, fungal morphology has the strong influence on the successful or unsuccessful production of the majority of fungal metabolites. Any actions to control fungal morphology should be made individually for the given species in conjunction with the metabolite produced by it, however the contemporary knowledge allows for certain generalisations. The variety of examples presented so far has proved that morphological engineering techniques are simple methods to control fungal morphology making it easier to deal with the problems exerted by the complicated growth pattern of the fungal species, even in the two species co-cultivations of filamentous microorganisms.

This work was financed from two projects funded by the National Science Centre (Poland): UMO-2015/19/B/ST8/02115 and 2017/27/B/NZ9/00534



OS - oral session

**The genome of the pathogenic white rot fungus *Armillaria ostoyae* encodes a distinctive genetic potential to degrade aromatic compounds**

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*Armillaria* species, representing white rot fungi with immensely enlarged subterranean colonies, are primary decay drivers in temperate forest ecosystems worldwide. Their ecological habits, besides the saprotrophic wood decomposing activities, include opportunistic invasive and necrotrophic interactions with woody hosts. Their complex lifestyle is maintained by an abundant spread of soil-borne rhizomorphs, which creates the potential for *Armillaria* species to encompass multiple hosts and to form extensive ecological networks.

To assess adaptive genomic and transcriptomic traits which may help *Armillaria* to conform to their host environment, we conducted comparative genomics, performed transcriptomics analysis on the morphogenesis of the rhizomorphs, and developed an in vitro stem invasion assay using the *Armillaria* – Norway spruce pathosystem.

Comparative genomics confirmed that *Armillaria* species have an extended potential in degrading pectins, aromatic compounds and to control proteins containing various chitin binding domains. Chitin and chitosan synthases, along with other genes involved in the biosynthesis and remodelling of the fungal cell wall, exhibited tissue-specific expression patterns in our morphogenesis experiment.

The in vitro assay, when combining freshly cut and autoclaved spruce tree stems, was found feasible to distinguish between virulent (C18) and non-virulent (C2) *Armillaria ostoyae* isolates as the virulent mycelia was more efficient in invading the cambium of the fresh stems. The differential gene expression analysis highlighted that all genes encoding phytotoxic ceratoplatanins and a significant pool of secreted enzymes involved in aromatic intradiol ring cleavage, were higher expressed in the virulent isolate under the fresh invasive conditions.

Our findings confirmed the adaptive invasive character of *Armillaria ostoyae* as a white rot fungus with a facultative necrotrophic lifestyle.



OS - oral session

**Lignicolous fungi – natural sources of ache enzyme inhibitors**

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**Background:** Constantly increasing interest in the study of bioactive substances of natural origin caused more intensive biochemical research of the macrofungi from which, in the last decades, have been isolated a large number of new compounds which exhibit significant biological effects such as polysaccharides, proteins, phenolics and terpenoids. Recently, fungi have also been recognized as producers of AChEIs, agents that inhibit the enzyme AChE from breaking down acetylcholine, thereby increasing both the level and duration of action of the neurotransmitter acetylcholine (ACh), hence can be used in the treatment of age-associated neurodegenerative diseases, such as Alzheimer.

**Objectives:** This study aimed to determinate at in vitro conditions anti-acetylcholinesterase activity of the selected extracts, EtOH, MeOH and H<sub>2</sub>O, derived from fruiting bodies of the lignicolous fungi, namely edible and non-edible species collected on the Fruska Gora mountain.

**Methods:** The bioactivity was estimated using Ellman's method at 412 nm during 5 min, while the results are expressed both as IC<sub>50</sub> and IC<sub>90</sub> values (50 and 90 % inhibitory concentration).

**Results:** The strongest inhibitory activity was observed for H<sub>2</sub>O and MeOH extract *C. disseminatus* (IC<sub>50</sub> = 0,034 µg/ml, IC<sub>90</sub> = 1.44 µg/ml), followed by *T. versicolor* EtOH (IC<sub>50</sub>= 0,47 µg/ml) and MeOH *A. aegerita*, *P. ostreatus* (IC<sub>50</sub>= 0,21 µg/ml, IC<sub>50</sub>= 0,50 µg/ml) and *G. sumneriana* (IC<sub>50</sub> = 1.0 µg/ml), while the lowest activity was detected for MeOH extract *P. ostreatus* (IC<sub>90</sub>=9,97 µg/ml). Further research should deepen the knowledge on biomedical potential of these lignicolous fungi in a regular diet of Alzheimer's patients.



OS - oral session

**Endophytic fungi improve Ni accumulation of hyperaccumulating plants**

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Endophytic fungi provide many benefits to the plant. Their popularity is growing, for their potential in agriculture and bioremediation. Agromining involves growing hyperaccumulator plants as a crop and processing it to recover target metals such as Ni without negative effects on the environment. Accumulation of Ni by the plant can be stimulated through the application of beneficial microorganisms, such as endophytes.

The aims of our investigations were: 1) to select appropriate endophytic fungi, which are able to improve biomass production and Ni extraction of hyperaccumulator plants and 2) to investigate the mechanisms of improved, fungus dependent Ni extraction. Fungal endophytes were isolated from hyperaccumulator plants *Noccaea goesingensis* and *N. caerulescens* inhabiting ultramafic soils in Europe. Several species of endophytes were found to be beneficial for the plant. The most promising endophytic fungi were Diaporthales and Pleosporales representatives. The endophyte increased Ni accumulation in its host *N. caerulescens*. In the non-host species, *N. goesingensis* affected Ni accumulation in an indirect manner by improving biomass production by the plant. The detection of endophyte in planta was performed by qPCR and a GFP-tagged fungal strain. It turned out that the mycelium developed inter- and intracellularly in root and shoot tissues within a few days after inoculation. Supplementation of the plant with the biostimulant increased nearly 5-fold fungal abundance in *N. goesingensis* leaves. Plants inoculated with Ni adapted endophyte showed higher root system development and lateral root elongation.

These results show the potential of endophytic fungi in agromining practices with the utilization of hyperaccumulator plants.

PS - poster session

**Characterization of minerals, elicitors in spent mushroom substrate extract and effects on growth, yield and the management *Cassava* mosaic diseases**

**Samuel Okere**<sup>1</sup>, Anthony Ataga<sup>2</sup>

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This paper evaluated the mineral compositions, disease resistance elicitors in *Pleurotus ostratus* and *Pleurotus tuber-regium* water extract spent mushroom substrate on the yield and management of cassava mosaic disease. The cassava plant let were generated through meristem tip culture before they were transferred to the screen house University of Port Harcourt Research Farm. The experiment was laid out in a completely randomized block design with 3 replicates. The data generated were subjected to analysis of variance (ANOVA). Means were separated using Fishers Least Significant Difference at P=0.05. The results obtained revealed that POWESMS contained 19.3, 0.52 and 0.1g/200g substrate of carbohydrate polymers, glyco proteins and lipid molecules elicitors respectively while it also contained 3.17, 212.1, 17.9, 21.8, 58.8 and 111.0 mg/100g substrate for N, P, K, Na, Mg and Ca respectively. Further PTWESMS contain 1.6, 0.04 and 0.2g/200g of the substrate as carbohydrate polymers, glycoprotein and lipid respectively; the minerals contained in this substrate were 3.4, 204.8, 8.9, 24.2, 32.2 and 105.5mg respectively for N, P, K, Na, and Ca. There were also significant differences in the mean values of number of storage roots, root length, fresh root weight, fresh weight plant biomass, root girth and whole plant dry biomass but no significant difference was recorded for harvest index. The result also revealed significant differences in mean values of disease severity index evaluated at 4, 8, 12, 16, 20, 24 and 28 weeks after inoculation.



PS - poster session

**Development of suitable substrate for *Sparassis latifolia* using beet pulp and corn flour**

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*Sparassis latifolia* has been known to contain a large amount of beta-glucan and are sold at high prices in the domestic market. However, the composition of the cultivation substrate for stable supply is currently insufficient. Due to these problems, contamination from long-term cultivation leads to high loss rate and low production efficiency. Because of this, it is difficult to expand cultivation farms in Korea. The cultivation experiment was carried out with 7 kinds of mixed-nutrient sources in order to select the cultivation medium for stable production. As a result of treatment with 20% by volume of each nutrient source, yields were 128.5g for GMSL69033 and 126.6g for 'Neoul' in substrate using beet pulp and corn flour(v/v, 15:5). At this time, the C/N ratio of the substrate was 107. The moisture content was 70% and the air filling porosity was 38%. In the future, We need to more study the relationship between chemical properties such as pH and C/N as well as physical properties such as volume expansion and maximum moisture content in substrate experiments.



PS - poster session

**The effect of different lignocellulosic substrates on the incubation and yield of oyster mushroom**

**Dušanka Bugarski**, Jelica Gvozdanović-Varga, Mirjana Vasić, Janko Červenski, Slobodan Vlajić  
*Institute of Field and Vegetables Crops, Novi Sad, Republic of Serbia*

Oyster mushroom is one of the best ecosystem cleaners. It plays an important role in the management of organic waste from existing surpluses of agricultural production, whose disposal began to be an environmental problem, and whose incineration unnecessarily wastes a large amount of organic materials. Therefore, special importance is given to research to obtain the highest quality substrate from the existing surpluses of agricultural production, from which the mushrooms will have as high a yield of mushrooms as soon as possible in the short incubation period.

In this study, the most frequent surpluses of agricultural production are soybean straw, maize stalks and sunflower husks, that were added as supplements to wheat straw, on which the oyster mushroom is most often produced in Serbia, in order to speed up the incubation period and increase yields for the three analyzed strain of *Pleurotus ostreatus*.

The yield obtained on pure soybean straw substrates, maize stalks and sunflower husks as well as their combinations with wheat straw was higher than the yield on the standard wheat straw substrate. It has been concluded from the results of the research that, in order to increase the yield of oyster mushroom, it is necessary to add supplements to the wheat straw, but that the rate of incubation is not correlated with the yield depending on the substrate.



PS - poster session

### **Biologically active compounds in lichenized fungi**

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Lichenized fungi (lichens) are unique symbiotic organisms that can live in habitats, which are too harsh or limited for most other organisms. As adaptations for life in specific habitats, lichens produce a large number of chemical compounds which possess numerous biological activities including antiviral, antibacterial, antitumor, etc. Thus, lichens may be of interest for pharmaceutical sciences and the industry itself.

The goal of the present research was to study the metabolites of lichens distributed in Israel. The different lichen species were collected in the Mediterranean territory of Israel. We investigated the composition and structure of the chemical compounds isolated from them. All studied species had a high diversity of lichen substances. Metabolites of two fruticose epiphytic species (*Ramalina lacera* and *Tornabea scutellifera*) and four foliose epilithic species from the genus *Collema* (*C. cristatum*, *C. callopismum*, *C. fuscovirens* and *C. flaccidum*) were studied in detail the most.

Four compounds belonging to monotetrahydrofuranic acetogenins have been isolated as new natural products from *Tornabea scutellifera*, and their biological activities were tested. The studied metabolites showed modest activity against different microorganisms and significant antitumor activity. Testing of biological activities of rare fatty acids isolated from *Ramalina lacera* and *Collema* species detected significant antibacterial and antifungal activities of these compounds. A photo protective mycosporine was found in calcicolous lichens *Collema cristatum* and *C. callopismum*. The study of this compound showed that it prevents UV-B induced cell destruction. Thus, our studies revealed that lichens distributed in Israel might be good potential sources of bioactive phytochemicals.

PS - poster session

**Wood-decay fungi studied under fermentative and oxygen-stress conditions**

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Reactive oxygen species (ROS) play an essential role in wood decomposition reactions and in the fungal decay mechanisms. White rot fungi decompose wood mainly via enzymatic reactions, by secreting carbohydrate-acting and lignin-attacking enzymes. In brown rot fungi wood decomposition occurs mainly non-enzymatically, via Fenton-chemistry reactions. In both decay types, hydrogen peroxide is needed for efficient decomposition of the wood and lignocellulose components. Under fermentative, oxygen-depleted conditions, however, the formation of ROS and hydrogen peroxide by fungi may be limited. Besides the fungal primary metabolism and metabolites, oxygen depletion and hypoxia conditions thus affect the wood decomposition processes and possibly, the production of fungal secondary metabolites. These compounds generally are biologically active and may act as redox mediators or ROS quenchers during the processes. However, a multitude of fungal bioactive compounds are unknown. Especially in brown rot fungi, it has been shown that specific secondary metabolites are involved in ROS mediated decomposition of the lignocellulose substrates but details of reactions during aerobic or anaerobic wood decay are still rather unknown. This has relevance also from an ecological point of view, since in nature, the decomposition conditions vary and may become partially oxygen limited. In this presentation, we review our current study on the topic and present results of our preliminary research of the effects of ROS addition on fungal decomposition of lignocellulose biomass under fermentative conditions.



PS - poster session

**CIRM-CF, a french Biological Resource Center dedicated to filamentous fungi for lignocellulosic biomass valorization**

**Delphine Chaduli**<sup>1</sup>, David Navarro<sup>1</sup>, Sabine Taussac<sup>1</sup>, Laurence Lesage-Meessen<sup>1</sup>, Chantal Parodi-Negri<sup>1</sup>, Sabine Genet<sup>1</sup>, Anne Favel<sup>2</sup>

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The CIRM-CF (Centre International de Ressources Microbiennes – Champignons Filamenteux) is a microbial Biological Resource Centre (BRC) dedicated to the preservation of filamentous fungi of interest for agro-industries. This young and growing collection organizes collecting operations in varying biotopes, including tropical regions. The majority of fungal strains isolated are naturally efficient to degrade lignocellulose and aromatic compounds. In 2018, the CIRM-CF collection gathers over 500 species of filamentous fungi and over 2,000 lignocellulolytic strains (60% belonging to Basidiomycota and 40% to Ascomycota).

Aiming high quality standards, the CIRM-CF is ISO9001 certified since 2006 for acquisition, authentication, conservation, diffusion and characterisation of its fungal resources. This BRC in conjunction with the BBF lab (Marseille, France), benefits from an exceptional scientific environment that allows the identification of strains using classical taxonomy, molecular tools and enzymatic characterization. These activities are also proposed to academic and industrial partners, to detect or identify fungi in different matrices, and to explore the enzymatic potential of fungal diversity by using a high-throughput screening platform.

We present three highlights that underline the benefits for research and development in biotechnology by screening well diversified and characterized microbial collections as mBRC (microbial Biological Resource Center).

The work was supported by the project NKFI K 124341 financed by the National Research Development and Innovation Fund.



PS - poster session

**Breeding and characteristics of new oak mushroom (*Lentinula edodes*) variety**

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Oak mushrooms (*Lentinula edodes*) mushrooms have traditionally been widely eaten in Asia and are one of the most popular mushrooms today due to their chewy texture and taste. The production of oak mushrooms in Korea is 23,983 tons in 2017, and its output is 218.1 billion won, which is the highest single item among mushrooms. Therefore, it is necessary to have good varieties of *Lentinula edodes*

'GMLE17522' was a new variety of *Lentinula edodes* developed by Mushroom Research Institute, GARES, Korea in 2017. It was bred by mating with monokaryons isolated from collected oak mushroom strains '36062-4' and 'LE15401-24'.

The new variety 'GMLE17522' takes 110 days for spawn running and browning, and 11 days for fruiting body developing, and the total cultivation period was 121 days. Characteristics of fruiting body of new variety 'GMLE17522' had brown hemispherical pileus, and the pileus and stipe were thicker than the control (Sanjo 701ho).

In the comparison of the yield (2 cycles cultivation) by 2.5kg sawdust substrates, the yield of new variety 'GMLE17522' was 416g, which was higher than 260g of control. In addition, the new variety '522' contains 79.4 mg / g of beta-glucan and 4.7 mg of polyphenol.

Therefore, the new varieties of mushrooms, which we have developed, are expected to increase farmers' income by improving mushroom productivity and quality.



PS - poster session

**The role of osmolytes and membrane lipids in the adaptation of extremophilic fungi to heat, cold and osmotic shocks**

**Vera Tereshina**<sup>1</sup>, Elena Ianutsevich<sup>1</sup>, Sofiya Bondarenko<sup>2</sup>, Olga Danilova<sup>1</sup>, Elena Bilanenko<sup>2</sup>

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The maintenance of the membrane functional state is a key element of the defense system under adverse conditions and can be accomplished both by changing the membrane lipid composition and with the help of osmolytes. It was shown that trehalose plays key role both in fungal thermophilia and alkaliphilia. The aim of this work is to study the role of osmolytes and membrane lipids in the adaptation of the thermophile *Rhizomucor miehei* and the alkaliphile *Sodiomyces tronii* to heat (HS), cold (CS) and osmotic (OS) shocks.

In *R. miehei* under optimal temperature conditions cytosol carbohydrates accounted for up to 12% w/w, and trehalose prevailed. As a result of CS and HS double decrease in trehalose level was observed. OS led to an increase of glycerol and arabitol levels. The characteristic feature of the lipid profile was the dominance of phosphatidic acids (PA) and phosphatidylethanolamines (PE). CS and HS led to an increase in the proportion of PA, while OS - to its decrease.

In *S. tronii* under optimal conditions the amount of soluble cytosol carbohydrates varied between 6-10% w/w. Trehalose and glucose dominated, while mannitol was present in small amounts. CS did not change the composition of carbohydrates, while HS doubled the level of trehalose. OS led to the sharp increase in the levels of arabitol and mannitol. Under the impact of CS and OS, there were no significant changes in the membrane lipids composition. After HS the proportion of PE increased, while PA - decreased.

Thus, despite high level of trehalose in the studied extremophiles, polyols are necessary for adaptation to OS, but not to CS and HS. The changes of membrane lipids were specific depending on the shock.

The study was supported by the Russian Foundation for Basic Research (project 18-04-00488).



PS - poster session

**Structure of the microbiome associated to commercial button mushroom cultivation**

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Mushroom cultivation consists of the development and fructification of fungal species in soil or selective substrates that provide nutrients and support for the crop. The microorganisms present in these environments strongly influence, and are required to generate the selective substrate and induce mushroom fructification. In this work, the fungal and bacterial microbiobiota living in different substrates employed in mushroom cultivation, compost (phase I, phase II and phase III, flush 1 and flush 2) and casing materials (day 1, day 6, day 8, flush 1 and flush 2), have been characterized by Next Generation Sequencing (NGS), analysis of phospholipid fatty acid content (PLFAs) and RT-qPCR. Additionally laccase activity and the content of complex carbohydrates in compost has been analyzed.

NGS through metagenomics provides a qualitative analysis of the non-cultivable microbiome while PLFAs and qPCR using specific primers are good techniques to quantify the microbial biomass in an ecological niche.

The bacterial diversity in compost and casing increased throughout the crop cycle together with the colonization of *Agaricus*. Phase I and phase II were dominated by microorganisms that show an active cellulolytic profile. *A. bisporus* showed a dominant role in phase III of the compost, displacing the pre-eminent Ascomycota. Interestingly the total bacterial biomass appears to be conditioned by the mycelium of the host, specially reflected by the PLFAs analysis of the phase III compost.

The techniques employed provide a reliable picture to determine the displacement of the microbial structure during the cultivation of mushrooms.

Project funded H2020 programme under the Marie Skłodowska-Curie grant agreement no. 742966 and also funded by the ADER project grant no. 2016-IDD-00013.



PS - poster session

**Prebiotic activity of hydrolysis product of (1→3)- $\alpha$ -D-glucans from fruiting bodies of *Laetiporus sulphureus***

**Paulina Adameczyk**<sup>1</sup>, Adam Waśko<sup>2</sup>, Małgorzata Pleszczyńska<sup>1</sup>, Katarzyna Próchniak<sup>1</sup>, Janusz Szczodrak<sup>1</sup>, Adrian Wiater<sup>1</sup>

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Prebiotics are defined as selectively fermented ingredient that results in specific changes in the composition and/or activity of the gastrointestinal microbiota, thus conferring benefits upon host health. Carbohydrates with prebiotics properties are obtained by direct extraction from fungi and plants or in a chemical or enzymatic process. One of the potential sources of the prebiotics are mushroom polysaccharides.

In this study water-insoluble (1→3)- $\alpha$ -D-glucans were isolated from fruiting bodies of *Laetiporus sulphureus*. To obtain oligosaccharides with beneficial impact on human health,  $\alpha$ -glucans were treated with sulphuric acid. Additionally, endo-(1→3)- $\alpha$ -glucanase was used to carry out enzymatic hydrolysis.

(1→3)- $\alpha$ -D-Glucooligosaccharides (GOS) obtained as hydrolysis products of (1→3)- $\alpha$ -D-glucans were tested for their prebiotic properties using lactic acid bacteria. Several *Lactobacillus* and *Bifidobacterium* strains were cultivated on MRS medium supplemented with GOS and standard prebiotics: fructooligosaccharides and inulin. The growth rate of probiotics was monitored by measuring optical density (OD 600) using a Bioscreen C instrument. The results showed that the ability of hydrolyzate of (1→3)- $\alpha$ -D-glucans to influence the growth of microorganisms was strain-selective. We observed three type of growth profiles for the lactobacilli and bifidobacteria when GOS was used as the sole carbon/energy source in MRS. Additional analysis showed that enteric bacteria do not growth in a medium supplemented with GOS.

Our results indicate that (1→3)- $\alpha$ -D-glucooligosaccharides obtained from (1→3)- $\alpha$ -D-glucans isolated from fruiting bodies of *Laetiporus sulphureus* possess prebiotic potential and might be used to modulate intestinal microbiota for health benefits.



PS - poster session

**Effect of pyrethroids on the phospholipid profile of *Beauveria bassiana***

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Contamination of the natural environment by insecticides is a worldwide ubiquitous problem. Entomopathogenic fungi, which possess the ability to attack and kill arthropods, are ecological alternative to chemical insecticides. These fungi can be used as mycoinsecticides instead of chemical insecticides. One of them, *Beauveria bassiana* is commonly applied as a bioinsecticide in many countries.

The aim of the study was to determine how pyrethroids, which are chemical insecticides, affect the structure and composition of the membrane of the entomopathogenic fungus *Beauveria bassiana*. The phospholipid profile and PC/PE ratio changes in the presence of pyrethroids:  $\lambda$ -cyhalothrin,  $\alpha$ -cypermethrin and deltamethrin in the concentrations of 5, 50, 100 and 200 mg/L were determined. Phospholipids were extracted using a chloroform-methanol mixture with the addition of glass beads. Afterwards, the extract was dissolved in 1 ml of methanol and then analyzed using the LC-MS/MS technique.

In *B. bassiana* biomass the highest content of phosphatidylcholines (PC) was found to be on the level of 60.33% of the total PL content. Phosphatidylethanolamine (PE), phosphatidylinositol (PI), phosphatidylserine (PS) and phosphatidic acid (PA) were on the level 21.24%, 10.34%, 7.16% and 0.93%, respectively. Statistically significant changes were observed in the content of PC in the cultures with the addition of  $\alpha$ -cypermethrin (100 mg/L) and deltamethrin (100 mg/L) and in the value of PE in cultures with the addition  $\lambda$ -cyhalothrin,  $\alpha$ -cypermethrin and deltamethrin (100 mg/L).

The obtained results suggest that pyrethroids in a high concentration of 100 mg/L adversely affect the membranes of *B. bassiana*.





PS - poster session

**Degradation of *Fusarium* mycotoxin zearalenone by *Isaria* sp.**

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Numerous fungal species have an ability to produce mycotoxins. Zearalenone (ZEN) is a product of secondary metabolism of *Fusarium* species whose natural habitat is soil, e.g. *F. graminearum*, *F. equiseti*, *F. culmorum*, *F. semitectum* or *F. incarnatum*. ZEN persists in the environment and is responsible for contamination of maize, oat, barley or wheat crops during growth, harvest and storage. Because of its similar chemical structure to estrogen, ZEN can affect reproductive disorders in cattle and chickens, infertility problems, especially in pigs and hyperestrogenic syndromes or immune disorders in humans. In view of toxicity of ZEN for living organisms, effective methods for its elimination from the environment require development.

The aim of the research was to define whether entomopathogenic fungi of the genus *Isaria* occurring in the soil like *Fusarium* sp. are able to degrade ZEN. Three strains were used in this study: *I. fumosorosea*, *I. tenuipes* and *I. farinosa*. Seven-day fungal cultures supplemented with ZEN at a concentration of 0.5 mg L<sup>-1</sup> were subjected to QuEChERS extraction. Liquid chromatography-mass spectrometry (LC-MS) technique was used to mark ZEN residue after seven days of cultivation. In all tested strains ZEN has not been detected, which means that *Isaria* sp. have the potential to 100% elimination of this mycotoxin. Additionally, two derivatives were determined in the *I. fumosorosea* post culture extracts. This strain transformed ZEN to a sulfonated derivative (397.2 m/z) and  $\alpha/\beta$ -zearalenol (319.6 m/z). Obtained results suggest possible interaction between ZEN producing fungi and *Isaria* sp.



PS - poster session

**In vitro testing of potential growth inhibitors of *Hymenoscyphus fraxineus***

**Miriám Kádasi Horáková**, Marek Kobza, Jozef Pažitný, Radovan Ostrovský, Marek Barta, Emília Ondrušková,  
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The pathogenicity of the ascomycete fungus *Hymenoscyphus fraxineus* as the major causal agent of ash dieback disease has been confirmed on *Fraxinus* spp. throughout Europe. In this study, an agar plate assay was carried out to assess sensitivity of *H. fraxineus* isolate to copper oxychloride, crystalline copper, sulphur, dodine, tebuconazole, zinc oxide, silver nanoparticles and gold nanoparticles for their potential to control the disease. 28 days after inoculation, copper oxychloride, sulphur and dodine showed the highest inhibitory effect (almost zero growth) on mycelial growth of the fungus. Tebuconazole, crystalline copper and zinc oxide inhibited in vitro growth by 68%, 47% and 30%, respectively, when compared to the control assay. The Ag and Au nanoparticles were classified as ineffective. The highly effective growth inhibitors (copper oxychloride, sulphur and dodine) were selected for use in tree injection experiments.

The study was financially supported by the Slovak VEGA grant agency under the project no. 2/0062/18.

PS - poster session

**Mechanical properties of thin films based on *Hericium erinaceus* mycelium**

**Maiia Ziangirova**<sup>1,3</sup>, Nailya Almyasheva<sup>1</sup>, Alexander Golyshkin<sup>1</sup>, Larisa Krasnopolskaya<sup>1</sup>, Andrey Teplykh<sup>2</sup>, Boris Zaitsev<sup>2</sup>, Irina Borodina<sup>2</sup>, Alexander Semyonov<sup>2</sup>, Vladimir Kolesov<sup>2</sup>, Iren Kuznetsova<sup>3</sup>

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It is possible to use films of higher fungi mycelium as sensor elements for gas acoustic sensors. The purpose of this work was to select the culture of basidiomycete, preparation of the film from biomass homogenizate and definition its mechanical properties. It was screened cultures of nine species of xylotrophic basidiomycetes. Mycelium of basidiomycetes was grown in submerged culture on a medium containing glucose, yeast extract and mineral salts. The culture *Hericium erinaceus* provided the highest yield of air-dry mycelium on the used medium (12g/l). A solvent in a ratio of 1:12 was added to the wet and washed with distilled water biomass and homogenized. The selection criterion of solvent was the uniform distribution of the homogenized mycelium in the liquid phase. The uniformity of distribution increased in the series: ethyl acetate<acetone<ethanol<water-ethanol mixture<water. Water was chosen as the solvent for the preparation of organic films. To obtain films with a thickness of 50–150 μm, 0.75–2.25 ml of homogenized mycelium was placed on the surface of one of the electrodes with diameter 22 mm of the CTBS-3 piezo-ceramic resonator and dried. The longitudinal (5–10 GPa) and shear (3–7 GPa) elastic moduli, viscosity (2–4 nPa\*s/rad) and density (200–500 kg/m<sup>3</sup>) of the films under study were defined by using the method of broadband acoustic spectroscopy. It was found that with decreasing film thickness, its elasticity and viscosity increase, and its density decreases. This variation can be explained by the fact that the surface of the film is rough, that leads to increased scattering of the acoustic wave at the free boundary of the film and increases the effective viscosity in the system.

This work was supported by the grant of the RSCF (Russia) No. 15-19-20046-P.



PS - poster session

**Evaluation of different *Trichoderma* species in their ability of degrading engine oil**

**Chiara Daccò<sup>1</sup>**, Marta Elisabetta Eleonora Temporiti<sup>1</sup>, Lidia Nicola<sup>1</sup>, Barbara Mannucci<sup>2</sup>, Federica Corana<sup>2</sup>, Solveig Tosi<sup>1</sup>

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Hydrocarbons can have harmful effects on organisms and environment, and conventional techniques for their removal are expensive, require the use of chemicals and long action times.

Bioremediation is the use of metabolic activities of microorganisms (algae, fungi and bacteria) to process hazardous substances in less toxic metabolites. *Trichoderma* is known to be active on different recalcitrant substrates, since it can produce a set of non-specific extracellular enzymes, generally involved in the degradation of lignocellulose rich substrata. However, the literature concerning the capacity of *Trichoderma* in the degradation of hydrocarbons is still limited.

The aim of this work was to investigate the capacities of *Trichoderma* to exploit used engine oil as the sole carbon source in the prospect of bioremediation of contaminated substrates.

Four different strains, isolated from different substrata and belonging to *T. asperellum*, *T. atroviride* and *T. harzianum*, were tested. As the first step, the fungi were evaluated for their ability of growing in biomass on polycyclic hydrocarbons and colorimetric tests were carried out to identify the production of specific enzymes, i.e. laccase and peroxidase. The strains were inoculated in a minimum liquid medium in direct contact with used engine oil for 45 days and samples were analysed by gas chromatography-mass spectrometry (GC-MS) every 15 days to detect which families of hydrocarbons had received a fungal attack.

The results showed that all the strains significantly changed the oil composition respect to the untreated control, and each strain had a different mode of action and efficacy in the biodegradation of used engine oil, suggesting the genus *Trichoderma* could be used as a bioremediation agent against hydrocarbons.

PS - poster session

**Management of *Fusarium* wilt by endophytic *Fusarium chlamydosporum***

**Vineet Meshram**, Meirav Elazar, Stanley Freeman

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Endophytic fungi are ubiquitous and polyphyletic groups of microorganisms that reside within the living tissues of their host plants without causing negative or detrimental effects. The endophyte–host relationships can provide benefits to the host, protecting it against biotic and abiotic stresses. *Fusarium* wilt of melon is caused by the soil borne fungus *Fusarium oxysporum* f. sp. *melonis* that results in plant mortality, reduced yield and impaired fruit quality. The present study focused on isolation and characterization of endophytic fungi from *Ecballium elaterium* collected from different locations of the Arava desert in Israel for the management of *Fusarium* wilt. A total of 130 endophytic fungi isolated from *E. elaterium* were screened to determine their antifungal potential against *F. oxysporum* f. sp. *melonis* (race 1.2) (FOM) using an in vitro antagonistic assay. Six isolates exhibiting inhibitory effect against FOM on quarter strength PDA plates, were selected for further studies. In growth chamber experiments, plants colonized separately with either spores or culture filtrates of the endophytic isolates EeS3 and EeR24 significantly protected plants from wilting as compared to water control and pathogen-challenged seedlings. Furthermore, in greenhouse experiments, plants colonized with EeS3 and EeR24v significantly improved and protected melon seedlings from wilting compared to those of the water control and pathogen-challenged treatments. Based on morphological and molecular phylogenetic analyses, both the endophytic isolates EeS3 and EeR24 were identified as belonging to the species *Fusarium chlamydosporum*. These results suggest that endophytic *F. chlamydosporum* may be developed as an effective biocontrol agent for the management of *Fusarium* wilt in melon plants.

PS - poster session

### **Rhizo-bioremediation of DDT-contaminated soils**

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Developing an inexpensive, effective and environmentally-friendly method of remediation of persistent organic pollutants is still an emerging need. Exploiting natural rhizosphere microorganisms, having capacity to degrade various xenobiotics, in association to plants able to accumulate the pollutants into their biomass, is a promising alternative to conventional remediation techniques. In this respect, fungi can play a key role.

Several reports are showing high potential of Cucurbitaceae in phytoremediation of DDT and its metabolites. In this study we present the results of rhizo-bioremediation of DDT using different varieties of *Cucurbita pepo*, inoculated in pot experiments with three fungal strains, isolated from DDT-contaminated soils in Poland, belonging to *Trichoderma*, *Rhizopus* and *Mortierella* genera. All the tested strains induced an increased uptake of DDT residues, either in case of “old” or recent contamination of the soil, in the *C. pepo* plants, thus reducing the content in the rhizospheric soil (about 50-60%) in comparison to control plants. The applied microfungi seemed also to influence the translocation of DDT from roots to the upper parts of the plant. Nevertheless, we did not detect DDT residues in the edible parts (fruit) of *C. pepo*. The proposed co-remediation strategy is discussed in view of possible interactions between plant genotypes and fungal strains that can affect the efficacy and/or mechanisms of this promising method of bioremediation.

The work was supported by a grant of the Polish Ministry of Agriculture and Rural Development.



PS - poster session

**Potential in mycoremediation of soil saprotrophic fungi: arsenic uptake and tolerance in different nutritional conditions**

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Arsenic (As) is a metalloid, naturally occurring in the earth crust. Anthropogenic sources, such as processing of arsenic-bearing minerals, are mainly responsible for environmental contamination, arising concern for toxic effects of As on human health and ecosystems. Fungi play key roles in fundamental biogeochemical cycles of essential and toxic elements and soil formation. Several studies reported that fungi can tolerate and accumulate high concentrations of arsenic and mediate processes of biovolatilisation through methylation. Our research aimed to evaluate As tolerance and bioaccumulation of four species of soil saprotrophic fungi and how nutritional conditions influence them. *Absidia spinosa*, *Purpureocillium lilacinum*, *Metarhizium marquandii* and *Cephalotrichum nanum*, previously isolated from areas with high natural As concentrations, were tested in two cultural media (MEA and Czapek-Dox). Results revealed high tolerance to all tested As concentrations (10, 20 and 50 mg/L sodium arsenite). Growth responses and tolerance to As have been investigated by tolerance indices (Rt:Rc (%) and T.I. (%)), based on growth data (diametric extension and dry weight). Most of these species resulted tolerant to all tested As concentrations, with growth responses also varying according to cultural media. pH medium after fungal growth was measured to study pH variation and metabolic responses. Chemical analyses by HG-AFS revealed As bioaccumulation. Elemental chemical composition of fungal biomass for all tested species was determined by ICP-MS and ICP-AES to assess how As occurrence and fungal responses to it influence the elements' uptake. Results shed further light on fungal geoactive roles in the environmental fate of As and provide potential applications in bioremediation.



PS - poster session

**Improvement of bottle cultivation method for automated shiitake (*Lentinula edodes*) cultivation**

Yeun Sug Jeong, Rhim Ryoo, Kang-Hyeon Ka, Hyo-rim Lee, Youngae Park, Yeongseon Jang  
*National Institute of Forest Science, Seoul, South Korea*

Shiitake (*Lentinula edodes*) is cultivated in Asia such as Korea, China, Japan, Vietnam and Taiwan. After the development of sawdust cultivation method, it has been replacing log cultivation method. Sawdust cultivation method is based on the usage of plastic bags, but they cause serious environmental problems. So, it is necessary to reduce plastic waste and while improving mushroom production efficiency and quality. We studied the possibility of applying bottle cultivation method for shiitake mushroom instead of sawdust bag method which was successfully applied to some edible mushrooms such as oyster mushroom. Three types of bottles were used in this study. When used large detachable bottles (2kg capacity), sawdust media showed lower weight loss rate and they needed longer incubation period than the other bottles. But small detachable bottles (400g capacity) could shorten incubation period and exhibit high yeild. So, this bottle was more suitable for bottle cultivation. Using this bottle, three incubation periods, two light conditions and two ventilation conditions were applied. As a result of this experiments, water spraying could shorten incubation period and advanced harvest time of the shiitake. However, in this bottle, the size of mushrooms decreased during the second harvest and total harvest period of mushrooms was shorter than other conditions. Under the same incubation period, high light intensity during incubation period and 55% air humidity by ventilation facility helps to improve the productivity. In this research, inhibition of lateral fruit body formation couldn't controlled completely, but, we could observe that shortening incubation period and reducing light transmittance to the lateral part of the media reduced lateral fruit body formation.





PS - poster session

**Fruiting-body formation of *Gymnopilus junonius* by Sawdust Cultivation**

Hyo-rim Lee, **Rhim Ryoo**, Yeun Sug Jeong, Yeongseon Jang, Kang-Hyeon Ka  
*National Institute of Forest Science, Seoul, South Korea*

*Gymnopilus junonius* is a poisonous mushroom belonging to the family Hymenogastraceae, and is found growing in dense clusters on stumps and logs of hardwoods and conifers. This species is stimulating the nervous system and causing hallucination symptoms. In addition, hispidin and bisnoryangonin of the chemical toxin reported from *G. junonius*. This study was conducted to research the cultivation condition of this species to develop its functional toxin material. For the cultivation of *G. junonius*, sawdust medium composed of *Quercus mongolica* and *Q. acutissima* with barely flours was used for the production of fruiting-body. The sawdust media measure ratio of 40% to 20% for a proper mixture. Each strain was cultured at  $23\pm 1$  °C and  $60\pm 5$  % humidity in the dark for 60 days and in the light for 30 days. After the mushroom primordia developed, the culture was moved to a facility maintained at  $18\pm 1$  °C and  $95\pm 5$  % humidity. In the further study, new crossed strain will develop using this strain of *G. junonius* and study new extracts material of this species to improve medicinal function.



PS - poster session

**Draft genome sequence and annotation of *Trichoderma hamatum* FBL 587  
(Sordariomycetes): insights into the mycoremediation of contaminated soil**

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Several *Trichoderma* species can synthesize molecules of high biotechnological value, including antifungal compounds and cell-wall degrading enzymes and provide applications in mycoremediation. The saprotrophic fungal strain FBL 587, deposited in the culture collection of the Fungal Biodiversity Laboratory, Sapienza University of Rome, was isolated from Polish DDT-contaminated soils. Tolerance indices (Rt:Rc (%); T.I. (%)) were used to assess fungal tolerance to 1 mg/L DDT. Metabolic responses to DDT through diametric extension, dry weight and medium pH after growth were also investigated, and results highlighted potential applications in DDT mycoremediation. DNA sequence analysis from the RPB2 and EF-1 $\alpha$  genes indicated that FBL 587 is closely related to *Trichoderma hamatum* (Bonord.) Bainer. To evaluate the safety of biotechnological application of this strain in DDT bioremediation, whole-genome sequencing (WGS), assembly and annotation were performed. In the genome sequence (38.9 Mb, GC 48.54 %), a total of 10,944 protein-coding genes were predicted and annotated with the GO and the CAZy databases. Bioinformatic analyses were conducted on genes and clusters of particular interest, including on putative loci involved in the metabolism of organochlorine and aromatic compounds. Prediction of biosynthesis gene cluster was also carried out, highlighting genes potentially involved in the production of compounds and secondary metabolites of biotech value. Additional research including RNA-Seq studies are ongoing to fully assess the potential for mycoremediation. The WGS project of *T. hamatum* FBL 587 has been submitted to GenBank (NCBI).

The costs for the Next Generation Sequencing (NGS) work for the WGS project were supported by INAIL-DIT, Rome (Italy).

PS - poster session

**Multiplex qPCR method for detection of *Verticillium* sp. and *Phytophthora* sp.**

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*Institute of Agrophysics, Polish Academy of Sciences, Lublin, Poland*

Increasing consumer awareness of fungicides used in conventional agriculture has caused organic farming to become more common in recent years. Organic farming methods contribute the crops to exposure to pathogens due to the exclusion of disease management by chemical sprayings. The abandonment of conventional fungicides creates the need for early and effective detection methods of causal agents of plant diseases. Fungal pathogens are serious threat causing large economic losses in food industry. Phytopathogens may impact the food production by reducing yield and decrease quality of harvest. *Phytophthora* sp. and *Verticillium* sp. are considered as common and threatening phytopathogens. Both of them are not host specific and there are no universal symptoms of disease on the plant.

Early detection of soil-borne diseases is very important for economic reasons. Traditional methods of fungal identification are time consuming and often inaccurate. However, emergence of molecular techniques such as quantitative polymerase chain reaction (qPCR) may overcome this problem. These techniques allow to detect the crucial pathogens with high precision, even before manifestation of the disease.

The aim of the study was to develop multiplex qPCR method for detection of *Phytophthora* sp. and *Verticillium* sp. in single reaction.

To achieve this goal, universal primers amplifying region D2 of Large Subunit of fungal rDNA, had been designed. Next, two hydrolysis fluorescent probes specific to the *Phytophthora* sp. and *Verticillium* sp. had also been developed.

This work was financed by The National Centre for Research and Development in frame of the project BIOSTRATEG, contract number BIOSTRATEG3/344433/16/NCBR/2018.

PS - poster session

**Chemicals released by fungi to overcome wood preservatives**

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Wood preservatives are of high importance for wood industry, as they improve the biological decay resistance of wood that is needed to apply wood outdoors for building uses. Due to the toxicity of most wood preservatives, increasing environmental awareness and more strict future chemical legislation, more sustainable wood preservative compounds need to be developed.

Research on new generation wood preservation chemical is nowadays focused in bio-based antifungals, such as tannins, pyrolytic liquids, natural oils or other biomass residues such as spent coffee often rich in constituents such as phenolics. However, some fungi are able to overcome the presence of phenolics or other antifungals. As an example, several fungi are able to produce melanin or oxidating hydroxyl radicals to fight the environmental stress via oxidative polymerization of phenolic compounds.

The present study will present inhibition caused by several bio-based chemicals against *Coniophora puteana* and *Trametes versicolor* and the production of chemicals by these fungi to overcome the presence of antifungals. To fulfil this aim, fungi will be grown in petri dish, with growth media amended with different bio-based chemicals. The results of this work will be relevant for identification of strategies used by fungi to detoxify treated wood, and thus, to develop better methods to formulate new generation wood preservatives.

PS - poster session

**Detection of *Fusarium* species producers of fumonisins in maize grains in Portugal**

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Worldwide, maize is among the most important food and feed crops. To ensure food quality, safety and security, maize should not be exposed to the risk of contamination by mycotoxins. The decrease of maize quality is mainly caused by fungal growth on ears and by their associated-mycotoxins production. The consumption of mycotoxin-contaminated maize results in acute or chronic consequences in human and animal health. For this reason, mycotoxin presence in maize grains is a current concern of maize producers and subject to marketing restrictions.

The most significant mycotoxins causing disease include aflatoxins, fumonisins, ochratoxin A, deoxynivalenol, zearalenone, and ergot alkaloids. Several species of fungi produce mycotoxins. The main producers belong to the genera *Aspergillus*, *Penicillium* and *Fusarium*. Fumonisins are mainly produced by *F. verticillioides* and *F. proliferatum*.

The aim of this study was to assess the effect of different crop management practices in *Fusarium* growth on ears and fumonisins presence in maize grains. Therefore, the identification and quantification of *Fusarium* species was determined in maize grains before and after storage. Different maize varieties grown under different nutritional regimens, water availability and soil-fungicide and insecticide applications were analyzed for two years.

The *Fusarium* spp. isolates were identified by macro and microscopic morphology and then confirmed by molecular tools. Results were correlated with the different management practices.

The study of *Fusarium* epidemiology in different management practices can be a key factor to reduce the fumonisins contamination in maize and prevent the risks for human and animal health by mycotoxins ingestion.

This work was supported by OG QualiMilho PDR2020, project n° 101-031295.



PS - poster session

**Mycelium growth of xylotrophic basidiomycetes on chemically modified lignocellulosic substrates**

Alexandr Golyskin, Nailya Almyasheva, Maria Yarina, Larissa Krasnopolskaya  
*Gause Institute of New Antibiotics, Moscow, Russia*

Bioconversion of lignocellulosic agricultural wastes is problematic due to their chemical structure and decomposition properties. Inhibiting components can be removed during pretreatment of lignocellulosic biomass by physical, chemical or biological means. Acid, alkali, peroxide pretreated and untreated pine and beech sawdust were evaluated as substrates for cultivation of *Hericium erinaceus* (Bull.) Persoon, *Flammulina velutipes* (Curtis) Singer and *Ganoderma lucidum* (Curtis) P. Karst. *G. lucidum* colonized both substrates containing alkali- and acid-pretreated sawdust. Acid pretreatment appeared to improve the bioavailability of lignocellulosic biomass for *F. velutipes* and *H. erinaceus*. Rise in the proportion of lignin led to an increase in growth rate of these basidiomycetes on substrates containing acid-pretreated sawdust compared to control. Peroxide and alkaline pretreatments did not show any significant effect on bioavailability of pine and beech sawdust for *F. velutipes* and *H. erinaceus*. The growth rate of *F. velutipes* on substrates with alkali-pretreated pine and beech sawdust was greater than the growth rate on control substrates, however the mycelium density decreased. No growth of *H. erinaceus* occurred on substrate containing alkali-pretreated pine sawdust.

Fruit bodies of *H. erinaceus* were grown on beech and pine sawdust with 10% wheat bran supplement. Dilute acid pretreatment of pine and beech sawdust reduced the time required for the initiation of fruiting of *H. erinaceus* from 65 to 35 days. Substrates containing pine sawdust produced *H. erinaceus* fruit bodies with higher phenolic contents than beech sawdust-based substrates.

PS - poster session

**The comparative study of phenolic metabolites produced by *Hericium erinaceus* and *Agrocybe aegerita* in vegetative and generative stages**

Nailya Almyasheva, Alexandr Golyshkin, Eugene Rogozhin, Larissa Krasnopolskaya  
*Gause Institute of New Antibiotics, Moscow, Russia*

Xylotrophic basidiomycetes produce a wide range of biologically active compounds including phenols, flavones and terpenoids with anti-inflammatory and antioxidant properties.

The aim of the present work is to study the antioxidant properties of secondary metabolites produced by *Hericium erinaceus* (Bull.) Persoon and *Agrocybe aegerita* (V. Brig.) at different stages of ontogenesis.

Vegetative mycelium of basidiomycetes were grown in submerged culture on a medium containing glucose, soybean meal and mineral salts. Fruit bodies of basidiomycetes were grown on beech/alder sawdust supplemented with wheat bran. Secondary metabolites were extracted from dried ground mycelium and fruit bodies with 80 % ethanol. To investigate the antioxidant properties of ethanolic extracts DPPH radical-scavenging activity assay, ferrous ions chelating assay and oleic acid peroxidation assay were applied.

The differences in composition and antioxidant properties of extracts of vegetative mycelium and fruit bodies of *H. erinaceus* and *A. aegerita* were observed. Ethanolic extracts of fruit bodies showed higher free radical-scavenging and chelating activity than the activity of vegetative mycelium extracts and completely inhibited the lipid peroxidation. Phenolic metabolites isolated from vegetative mycelium of *H. erinaceus* were found to decompose hydroperoxides. Components of ethanolic extracts were separated on reverse-phase C18 HPLC column. The unknown compounds were identified by HPLC coupled with mass spectrometry (HPLC/MS).



PS - poster session

**Cultivation of *Ganoderma lucidum* using residues from forest industries**

**Marta Cortina-Escribano**, Pyry Veteli, Riikka Linnakoski, Henri Vanhanen  
*Natural Resources Institute Finland, Joensuu, Finland*

Forest industries generate low-value side streams that could be suitable for the cultivation of edible and medicinal mushrooms. *Ganoderma lucidum* (W. Curtis) P. Karst (Basidiomycota) is a well-known species that contains several bioactive compounds, which define the market value of the fungus. The aim of this study was to collect information about suitability of substrate media containing various wood residues for the cultivation of *G. lucidum*, as well as compare strain performance.

*Betula pendula*, *Populus tremula*, *Larix* sp., *Pinus sylvestris*, *Alnus incana* and *Picea abies* sawdust and wood chips were used as substrates. Several strains of *G. lucidum* originating from Finland were tested. The substrate bags (1kg dry weight) were filled with 60% of wood chips and 40% of sawdust, except for *B. pendula*, in which case 100% wood chips were used. Five replicates per each substrate type were inoculated with 150 ml of barley spawn. When the substrate bags were completely colonized, they were kept at 26 °C with a relative humidity of 80% to support fruiting body formation. Observations on the yield and substrate mass loss were annotated.

The results suggest that the most suitable wood residue type for the cultivation of *G. lucidum* is *B. pendula* followed by *P. tremula*. Most of the strains had difficulties developing fruiting bodies on the substrate media containing coniferous wood (*P. sylvestris*, *Larix* sp. and *P. abies*). These findings demonstrate the importance of substrate media and strain selection for mushroom cultivation purposes.



PS - poster session

**Ascomycota composition based on next generation sequencing in soil amended with spent mushroom substrate**

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Spent mushroom substrate (SMS) is composted organic material remaining after harvesting of mushroom crop. The SMS consists of agricultural residues with the addition of poultry manure, coal, peat and other components. SMS has a high content of organic matter, nitrogen and phosphorus and low levels of potassium, calcium and magnesium.

The goal of the study was to determine Ascomycota composition in long-term amended soil with spent mushroom substrate. The next generation sequencing (NGS) through Illumina MiSeq technology was used to determine soil fungal diversity based on the ITS1 marker. The FUNGuild online application was used to assign functional information to OTUs in high-throughput sequencing datasets.

Based on the total high-quality sequences, Sordariomycetes, Eurotiomycetes and Pezizomycetes were the most dominant classes in SMS-treated soil, accounting for 47%, 8% and 8%, respectively. Moreover about 30% of obtained sequences were unidentified. Taking into account trophic group of fungi, the results showed that the fungal community was dominated by saprotroph (53%), pathotroph-saprotroph-symbiotroph (30%) and saprotroph-symbiotroph (10%). In the saprotroph group most functional guilds was unidentified (37%) and the rest was represented by soil, wood and dung saprotroph. The guild of pathotroph-saprotroph-symbiotroph was dominated by animal pathogen-dung, saprotroph-endophyte-epiphyte-plant, saprotroph-wood saprotroph and the rest was represented by endophyte-plant, parasite-plant and pathogen-wood saprotroph.

The study was partially funded by HORIZON 2020, European Commission, Programme H2020-SFS-2015-2: Soil Care for profitable and sustainable crop production in Europe, project No. 677407 (SoilCare, 2016-2021).



PS - poster session

***Fusarium temperatum* – the important producer of beauvericin and enniatins**

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*Fusarium temperatum* Scaufl. & Munaut is considered as a one of the most important causing factor of maize ear rot in Poland. The fungus produces two main secondary metabolites, like beauvericin (BEA) and the structurally related enniatins (ENN). The aim of studies conducted in 2018 was analysis of the profile of BEA and ENN derivatives, FB1, FB2 and ERG produced by *F. temperatum* in laboratory conditions. Biosynthesis of secondary metabolites was performed in vitro on rice and maize, according to methods described by Waśkiewicz et al. (2012). Beauvericin, enniatins (ENN A1, ENN A, ENN B, ENN B1), fumonisins (FB1, FB2) and ergosterol (ERG) were extracted using methanol and analysed by HPLC. The mycotoxin profile of 70 *F. temperatum* strains, originally collected from naturally infected ears has been studied. BEA was recognised as predominate hexadepsipeptide toxin. The mean level of BEA was 812.01 ppm on rice and 140.98 ppm on maize kernels. On rice the mean level of: ENN A1 was 37.09 ppm, ENN A - 5.27 ppm, ENN B - 3.51 ppm and ENN B1 - 48.81 ppm. On maize kernels all of tested isolates of *F. temperatum* produced ENN A1. Among all 70 isolates, 17 strains of fungus synthesized ENN A (mean 24.74 ppm), 18 isolates produced ENN B (mean 72.74 ppm) and 11 isolates produced ENN B1 (mean 30.84 ppm). Trace amounts of fumonisin FB1 on rice and maize kernels were noted for 5 isolates of fungus respectively. On rice the ERG level ranged from 15.70 ppm to 428.10 ppm. On the second maize kernels used as substrate, the ability of producing ERG was significantly higher. The mean level of ERG was 181.22 ppm (on rice) and 282.28 ppm (on maize kernels). Quite scant and contradictory reports in the literature about spectrum of *F. temperatum* mycotoxin production points to the need continuous in this aspect study.

Acknowledgements Part of the study was supported by Ministry of Agriculture and Rural Development Task Number 92.

## FUNGAL INTERACTIONS



OS - oral session

**Interactions of polyporoid fungi affect enzyme activities, release of volatile organic compounds and wood decomposition pattern**

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In terrestrial ecosystems, forests are significant carbon sinks. Forest dead wood is a carbon storage as well as habitat for animals, microbes, and fungi that display saprotrophic, pathogenic, or ectomycorrhizal lifestyle. Fungal communities in dead wood are dynamic with interactions deviating from mutualistic or neutral to antagonistic in character. Especially the antagonistic reactions are visible in hyphal confrontations.

We studied the influence of *Fomitopsis pinicola*, a common brown rot Basidiomycota Agaricomycetes species encountered in boreal and temperate forests as a saprotroph and tree pathogen, on hyphal growth and enzyme activities in the presence of five white rot species including *Phlebia radiata* and *Trichaptum abietinum*. The fungi were cultivated in various species combinations under controlled conditions on agar media, wood-supplemented medium and on solid spruce wood. In fungal co-cultures, *F. pinicola* was a supreme colonizer advancing over hyphae of the white rot species whereas with *P. radiata*, the other white rot species were provoked leading to formation of dense mycelial fronts. Substantial production of oxalic acid by *F. pinicola* occurred in liquid and solid-state wood cultures, and evidence of brown rot Fenton reaction biochemistry was also obtained. Emitted volatile organic compounds from the fungal cultures on spruce wood demonstrated a distinct effect and VOC profile generated by white rot compared to brown rot decomposition of spruce wood.

In conclusion, fungal interactions affected enzyme and chemical activity dynamics over time resulting with different patterns of wood decomposition, release of volatile and dissolved carbon compounds, and recycling of organic nitrogen. We aim at extending the experiments to forest site studies.

OS - oral session

**Compositional shift in communities of bacteria and fungi following tree species change from natural birch to planted spruce forests**

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Interaction between above- and below-ground biota are fundamental drivers of ecosystem processes; essentially mediated through the microorganisms; which are susceptible to both vegetation and soil environment changes. All over the world, non-native tree species are introduced and established in plantations for wood production. In western Norway, spruce have been planted for over 50 years, replacing the native birch forests. We investigated the impact of this tree species shift on the belowground microbial communities and fungal biomass, as well as their relationship with the carbon stock, across the different soil layers. We observed significant influence of tree species shift on the bacterial and fungal diversity, both being lower in the spruce forests. Shift in tree species had stronger impact on fungal communities compared to bacteria, and was more pronounced so in the upper forest floor layers. Ergosterol level was similar in mineral layers of both forest types, but significantly higher in organic layer of spruce forest, a pattern similar to C:N ratio. The effect of trees species on the bacterial and fungal communities were related to variation in understory vegetation biomass. Abundance of ectomycorrhizal fungi were relatively higher in the spruce forests and vice versa for saprotrophic fungi. In the spruce forests, the dominance of ectomycorrhizal fungi increased and saprotrophic fungi decreased with soil depth, a trend absent in the birch forests. The shift in tree species, is accompanied by a shift in microbial communities and fungal biomass with tentative impacts on C sequestration processes.



OS - oral session

**Biology, epidemiology and host range of the downy mildew of sage (*Peronospora salviae-officinalis*)**

**Mascha Hoffmeister**<sup>1</sup>, Wolfgang Maier<sup>1</sup>, Marco Thines<sup>2</sup>

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*Salvia officinalis* is an ancient medicinal plant, and by far the economically most important sage species. Since its first report in 1993 a new emerging disease has been reported from all over the world and is of increasing economical concern. The causal agent of the disease was described as a species of its own in 2009. Nevertheless, very little is known about its infection biology and epidemiology. The rapid worldwide spread of *P. salviae-officinalis* through sage cultivation and own observations from sage stands in Germany suggested that this downy mildew could be transmitted via contaminated seeds, but neither seed contamination nor oospore production had been proven for *P. salviae-officinalis*. The aims of the current study were therefore to unravel the life cycle of this downy mildew and gain deeper insights into the epidemiology of the disease. Optimal climate conditions for infection and sporulation were assessed by artificial inoculations in climate chambers. Using confocal laser scanning microscopy and light microscopy we tracked the whole life cycle of *P. salviae-officinalis* including oospore formation. Contamination of sage seeds by *P. salviae-officinalis* was also proven by seed washing and by PCR and DNA sequence comparisons. In addition, molecular phylogenetic analyses of downy mildews from other sage species suggested that *S. sclarea* is also infected by *P. salvia-officinalis*, and could play a role as an additional host, while the downy mildew on *S. pratensis* belongs to a different species.



OS - oral session

**Competitive interactions between the plant pathogen *Armillaria mellea* and root endophytic *Trichoderma* spp. or the opportunist *A. gallica***

**Jassy Drakulic<sup>1</sup>**, Helen Rees<sup>2</sup>, Matthew Cromey<sup>1</sup>

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*Armillaria* root rot (ARR) is a destructive disease of a broad range of plants. It has been the disease most frequently diagnosed by the RHS Gardening Advice Service for more than two decades. A four-year survey of diseased garden plants showed that ARR is caused predominantly by the pathogenic species *Armillaria mellea*, but 15% of infections were by the opportunist *A. gallica*. Presently, disease is not diagnosed to species level. Advice is based on the assumption that pathogenic *A. mellea* is present. Disease control is costly and labour intensive, and ARR has a high rate of reoccurrence.

One research direction looked for beneficial microbes to help young plants evade ARR in areas with previous infection. Roots were sampled from susceptible plants that had survived in proximity to infections at RHS Wisley. *Trichoderma* spp. endophytes were isolated and competition studies were performed in vitro and in planta. In dual culture, *Trichoderma* isolates rapidly grew over *A. mellea*, which was killed after six weeks. Seven of 40 isolates initially screened in *Fragaria* showed potential to protect against ARR. These seven isolates were further tested in *Ligustrum* to show their efficacy in a woody host, along with evidence of their retention within host roots.

A second direction explores the hypothesis that *A. mellea* is excluded by *A. gallica*. *Ligustrum* plants were inoculated first with *A. gallica* then with *A. mellea* after three or six months. By 18 months post inoculation the highest mortality was in control plants with only *A. mellea* added. The lowest mortality was when *A. mellea* was added six months after *A. gallica*. A repeat was prepared using increased replication, an improved model host system, and quantification of infection of each species using new QPCR primers.



OS - oral session

**The impact of Southern Pine bark beetle on the decomposition and fungal success of pitch pine wood in the NJ pine barrens**

**John Dighton**<sup>1</sup>, Ning Zhang<sup>2</sup>, Emily Walsh<sup>2</sup>

<sup>1</sup>*Rutgers University Pinelands Field Station, New Lisbon, USA*

<sup>2</sup>*Rutgers University, New Brunswick, USA*

With a change in climate patterns, warmer winters in New Jersey, USA have allowed Southern pine bark beetles to invade pines in the NJ pine barrens. The pine barrens is an ecologically unique and protected area in New Jersey, so the impacts of bark beetles in this system is of great concern for conservation. Since bark beetles in general import the fungi they depend on to help feed their larvae, we are addressing three aspects of introduction of fungi into wood, which will ultimately join the decomposer pool as beetles kill their host tree. We look at (i) does the introduction of beetle fungi affect the rate of wood decay, (ii) does this unique fungal introduction influence the subsequent succession of decomposer fungi into this decaying wood and (iii) does the natural tree defense response to damage, of resin production influence the rate of wood decomposition and the succession of fungi effecting that decomposition. Wood and bark invaded by beetles has a slower decomposition rate than wood and bark not influenced by beetles. Using community DNA analysis, there are significant differences in the fungal community developing on decomposing beetle infested wood and bark to un-infested material. The presence of resin significantly decreases wood decomposition.



OS - oral session

### **Large-scale controls of ectomycorrhizal fungi**

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Explaining the large-scale diversity of soil organisms that drive biogeochemical processes — and their responses to environmental change — is critical in our rapidly changing planet. However, identifying consistent drivers of belowground diversity and abundance for functionally key soil organisms at large spatial scales has remained problematic.

We investigated a major guild, the ectomycorrhizal fungi, across European forests at unprecedented spatial scale and resolution and with in situ data, to determine key biotic and abiotic predictors of ectomycorrhizal diversity and to identify dominant responses and thresholds for change across complex environmental gradients.

We showed the effect of host, environment, climate and geographical variables on ectomycorrhizal diversity, and defined thresholds of community change for key variables. We quantified specificity to tree hosts and revealed plasticity in functional traits involved in soil foraging across gradients.

We conclude that environmental and host factors explain most of the variation in ectomycorrhizal diversity, that the environmental thresholds used as major ecosystem assessment tools (i.e. critical loads) need strong downward adjustment to protect ectomycorrhizal fungi, and that the importance of belowground specificity and plasticity has previously been underappreciated. This research is a fundamental platform to achieve large-scale long-term biological monitoring of forest soils by providing a first robust benchmark.



OS - oral session

**Endophytic fungi improve Ni accumulation of hyperaccumulating plants**

**Rafał Ważny**<sup>1</sup>, Piotr Rozpądek<sup>1</sup>, Agnieszka Domka<sup>1</sup>, Roman J. Jędrzejczyk<sup>1</sup>, Katarzyna Turnau<sup>2</sup>

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Endophytic fungi provide many benefits to the plant. Their popularity is growing, for their potential in agriculture and bioremediation. Agromining involves growing hyperaccumulator plants as a crop and processing it to recover target metals such as Ni without negative effects on the environment. Accumulation of Ni by the plant can be stimulated through the application of beneficial microorganisms, such as endophytes.

The aims of our investigations were: 1) to select appropriate endophytic fungi, which are able to improve biomass production and Ni extraction of hyperaccumulator plants and 2) to investigate the mechanisms of improved, fungus dependent Ni extraction. Fungal endophytes were isolated from hyperaccumulator plants *Noccaea goesingensis* and *N. caerulescens* inhabiting ultramafic soils in Europe. Several species of endophytes were found to be beneficial for the plant. The most promising endophytic fungi were Diaporthales and Pleosporales representatives. The endophyte increased Ni accumulation in its host *N. caerulescens*. In the non-host species, *N. goesingensis* affected Ni accumulation in an indirect manner by improving biomass production by the plant. The detection of endophyte in planta was performed by qPCR and a GFP-tagged fungal strain. It turned out that the mycelium developed inter- and intracellularly in root and shoot tissues within a few days after inoculation. Supplementation of the plant with the biostimulant increased nearly 5-fold fungal abundance in *N. goesingensis* leaves. Plants inoculated with Ni adapted endophyte showed higher root system development and lateral root elongation.

These results show the potential of endophytic fungi in agromining practices with the utilization of hyperaccumulator plants.

PS - poster session

**Monitoring without touching: can biocalorimetry reveal different fungal life history strategies and fungal interactions on solid substrates?**

Hieu Linh Duong, Sven Paufler, Thomas Maskow, Dietmar Schlosser

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Fungi employ different life history strategies for substrate utilization. R-selected fungi quickly colonize new resources and preferentially degrade easily available compounds. They rapidly collapse in response to nutrient depletion. By contrast, slow-growing C strategists utilize even complex recalcitrant substrates with the help of certain functional attributes. During the simultaneous colonization of a substrate fungi may interact in different ways, depending on the combination of species involved.

We have applied biocalorimetry in order to record fungal activities and interactions during growth on a solid lignocellulosic substrate (wheat straw), and aimed to link the related data to different life history strategies. To this end, we measured and compared the metabolic heat production rate generated by the litter-decay basidiomycete *Stropharia rugosoannulata* (C behaviour), the ubiquitous ascomycetous mold *Stachybotrys chlorohalonata* (presumable R-strategist), and a mixture of these two fungi during growth on wheat straw.

In accordance with a R-driven strategy, calorimetric data demonstrated that the growth-related activity of *S. chlorohalonata* rapidly increased during the initial phase of the experiment and was highest already at culture day 4. *Stropharia rugosoannulata* started growing after 4 days of incubation and growth remained at a rather constant rate thereafter, in line with the behaviour of a C strategist. The growth-related activity of a co-culture of both fungi rather resembled the activity pattern of *S. chlorohalonata* than that of *Stropharia rugosoannulata*.

Briefly, real-time biocalorimetry holds promise as a rapid screening tool for studying different fungal life history strategies and interactions during growth on solid lignocellulosic substrates.

PS - poster session

**Biodegradation of single use plastics by white-rot fungi**

Denise Hassinger<sup>1</sup>, **John Dighton**<sup>2</sup>

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Plastic pollution is a recognized global threat that must be resolved in order to preserve and conserve natural environments and the animals within them. White-rot fungi possess lignolytic enzymes which may be important for the decomposition of plastics. We selected five fungi that are grown commercially (*Pleurotus ostreatus*, *P. ostreatus columbinus*, *Lentinula edodes*, *Ganoderma lucidum*, and *Trametes versicolor*) and tested their ability to break down single-use plastics used in the catering industry (high-density polyethylene (HDPE), low-density polyethylene (LDPE), polypropylene (PP), polystyrene (PP), and polyethylene terephthalate (PET)). We found a diversity of efficiencies in decomposition between fungi and between plastics. Mass loss of plastic was determined in cultures of fungi on tap water agar, 25% MEA or lignin agar after one or three months incubation. High-density polyethylene (HDPE) was more susceptible to fungal degradation than low-density polyethylene (LDPE). Fungal species showed significant differences in their ability to degrade HDPE. *Pleurotus ostreatus*, *P. ostreatus columbinus*, *Lentinula edodes* displayed the highest biodegradation abilities of HDPE, and performed the best in the lignin media, where it is suggested that the presence of lignin stimulated the production of enzymes that degrade plastics. The plastics PP, PS and PET, have been shown to be highly recalcitrant in nature, so it is unsurprising that minimal mass loss was obtained after one- or three-month incubation with the fungi.



PS - poster session

**The effect of abiotic factors on growth of ophiostomatal fungi - Factors shaping microbial communities of ecologically important bark beetles**

**Karel Švec**

*Institute of Microbiology of the CAS, v. v. i., Prague, Czech Republic*

Barkbeetles (Scolytinae) belongs between most diverse groups of insect. This group have global socio-economical impact. Symbiosis of barkbeetles and microorganisms allows them to reach new sources of nutrients, extend their niches, defend themselves against their predators and adapt to fast changing conditions. In my PhD thesis I focused mainly on the abiotic factors like effect of oxygen, pH or activity of water on growth of ophiostomatal fungi. Basic abiotic factors are considered to be responsible for division into 2 main groups of barkbeetles (co called „ophiostomatoid fungi associated“ and „*Geosmithia* associated“ types) according to our hypothesis.

PS - poster session

**Mycorrhizal fungi modulate nitrate inhibitory effect on *Dactylorhiza majalis* seed germination**

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Many orchid species are currently endangered, some of them disappearing with no obvious reason. Very little is known about their specific physiology including the regulation of seed germination. Mature seeds of several terrestrial species do not germinate in vitro making their cultivation for scientific and rescue purposes impossible. As we have shown previously, nitrates are able to strongly inhibit asymbiotic orchid seed germination. Here, we tested whether orchid seed germination is sensitive to nitrates also in the presence of mycorrhizal fungi. We germinated *Dactylorhiza majalis* symbiotically, on cultivation medium containing different concentrations of nitrates. Two compatible strains of *Ceratobasidium*, two of *Tulasnella* and one *Sebacina* were tested. Germination was negatively influenced by nitrate similarly as on asymbiotic culture media with 4 out of 5 fungal strains. Surprisingly, one *Ceratobasidium* strain, caused insensitivity of seeds to nitrate so germination rate was unaffected by nitrate concentration. Further growth of protocorms was not affected by nitrates. Both germination and further growth of protocorms also differed between each fungal strain. These results suggest that various mycorrhizal fungi can exhibit different effects on orchid. We also showed that nitrates can strongly inhibit symbiotic germination which supports the hypothesis that recently elevating nitrogen levels in nature might be responsible for recent decrease in number of European orchid sites.

PS - poster session

**Role of the microbial symbionts in the bark beetle holobiont**

Tereza Veselská, Karel Švec, Martin Kostovčík, Miroslav Kolařík, Paula García Fraile  
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Bark beetles (Coleoptera: Curculionidae) are ecologically and economically significant herbivores. Phloeophagous bark beetles primarily feed on plant tissues; nevertheless they are associated with broad range of microorganisms, creating a holobiont more or less specific to beetle. The holobiont, including mutualistic, antagonistic or commensal fungi, bacteria, mites and nematodes, has functional importance to symbiosis. The core symbionts probably improve beetle fitness by aiding with diet, detoxification of plant tissue and suppressing growth of pathogens. Although most bark beetles are attracted by weakened trees, they can become aggressive tree-killing pest during their outbreaks. Among the European species, *Ips typographus* is surely the most important pathogen of spruce, responsible for huge economic losses. The fungal associates have been suggested to contribute to tree killing, e.g. the common fungal symbionts of *I. typographus*, *Endoconidiophora polonica* and *Ophiostoma bicolor*, can detoxify tree defensive compounds. However, little is known about further functions of associated microorganisms and their effects on *I. typographus* fitness and population dynamics. The aims of our study are to describe the composition and functions of the symbiont community of *Ips typographus* and to identify the core symbionts. Firstly, we will study the composition of holobiont community using the metagenomics approach in different stages of beetle development. Secondly, we will identify the main metabolic paths presented in *I. typographus* system by metatranscriptomics approach. Then, we will assess whether these functions are supported by multiple associates. Moreover, based on in situ hybridization, we shall localize the endosymbionts within the beetle body.



PS - poster session

**Metagenomic analysis of fungi and bacteria in rhizosphere soils around four basidiomycete mushrooms in Jeju Island in Korea**

Dong Hyeung Lee<sup>1</sup>, Hyung Jin Noh<sup>1</sup>, Kang Hyo Lee<sup>2</sup>, **Seong Hwan Kim**<sup>1</sup>

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Gothawal in Jeju Island, a volcanic island in Korea is considered an internationally important wetland. Thus, understanding of mushroom diversity in this area is one of interesting research topics. We are interested in microbial community of rhizosphere soil of mushroom which could interact with the mushroom. Operational taxonomic unit(OTUs) analysis of ITS and 16s rDNA V3V4 regions was carried out to obtain basic information on the fungal and bacterial community present in *Amanita caesarea*, *A. verna*, *Cortinarius violaceus*, and *Laccaria vinaceoavellanea*. The most diverse of OTUs were found in the rhizosphere soil of *A. verna*, which had 1259/4345 OTUs(ITS/V3V4). And then 1013/3918 OTUs(ITS/V3V4) from *L. vinaceoavellanea*, 900/3001 OTUs(ITS/V3V4) and 625/2899 OTUs(ITS/V3V4) were analyzed from *A. caesarea* and *C. violaceus*, respectively. Species richness was considerably higher in the rhizosphere soil of *A. verna* and *L. vinaceoavellanea* than that of others. The most abundant bacterial phylum was Proteobacteria, followed by Acidbacteria, while Bacteroidetes were dominant in the rhizosphere soil of *A. verna*. Interestingly, the most abundant fungal phylum in the four rhizosphere soils was Basidiomycota, followed by Ascomycota and Mortierellomycota. At the level of genus, the dominant fungal group differed among the four rhizosphere soils. Among the isolated bacteria from four rhizosphere soils, *Bacillus laterosporus*, *B. velezensis*, *B. laterosporus*, *K. kifunensis*, *L. xylanilyticus*, and *P. tibetensis* strongly inhibited the mycelial growth of the kinds of mushroom species in the co-cultivation experiment.





PS - poster session

**Interactions in contaminated environment: Investigation of *Schizophyllum commune* in Chernobyl soil**

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Radionuclide pollution threatens environments, e.g. after the nuclear power plant melt down that occurred April 26, 1986 in Chernobyl, Ukraine. There, approximately  $3 \times 10^{15}$  Bq were released into the environment. Even after more than 30 years, thousands of acres of land in the exclusion zone are still highly contaminated, especially with cesium.

Since fungi show high radio-tolerance and can accumulate radionuclides as well as heavy metal stable isotopes, they may present good means for bioremediation of the contaminated soil. To investigate the potential of radionuclide protection, the white rot fungus *Schizophyllum commune* and the saprotrophic fungus *Leucoagaricus naucinus* were inoculated to a test field with crop plants established at a distance of just 10 km from reactor block 4 at the Chernobyl site.

Therefore, the soil was analyzed for its element content and for microbial community. Using species specific primers, *S. commune* was shown to survive over a time of at least 12 month in the soil on site and at least 6 month on water agar with Chernobyl soil. This was unexpected, since *S. commune* is thought to be specifically wood-colonizing. In contrast, the saprotrophic *L. naucinus* was not re-identified, even though for this fungus, meadows are the natural habitat.

Additionally, an experimental set-up was created which enables *S. commune* to grow in soil under laboratory conditions to allow for comparison between different soils and growth media. Here, RNA-seq was performed to identify tolerance mechanisms. With RNA extracted from two soils with different heavy metal contents, as well as from liquid media with or without heavy metals, underlying mechanisms could be deduced to educate further, more specific experiments.

PS - poster session

**Evaluation of the secondary metabolism of the genus *Laccaria* (Agaricomycetes, Hydangiaceae)**

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*Laccaria* is a cosmopolitan genus of facultative ectomycorrhizal (ECM) Basidiomycota associated with numerous tree species.<sup>[1]</sup> Moreover, some taxa of the genus were shown to be “pioneer fungi”, highly beneficial for their hosts.<sup>[1,2]</sup> Due to the ability to form stable cultures in vitro, *Laccaria* species, and in particular, *L. bicolor*, the first ECM fungal genome sequenced,<sup>[3]</sup> have been used as important experimental model organisms for decades. However, while extensive research focusing on the ecology and genetics of *Laccaria* has been conducted, the secondary metabolites of the genus widely remained to be explored.

The first comprehensive mycochemical study on the basidiomes of some *Laccaria* species have recently led to the discovery of several previously unknown secondary metabolites with interesting molecular structures and biological activities.<sup>[4]</sup> Moreover, metabolic profiling indicates the presence of potential chemotaxonomic markers that might solve the existing problems in the taxonomy of some *Laccaria* species.<sup>[4b]</sup> The isolated secondary metabolites belong to different classes like 3-acyltetramic acids, anthranilic acid derivatives, and 1,3-diazacyclooct-1-ene alkaloids, showing that the fungi are capable of expressing a wide array of secondary metabolite biosynthesis pathways. These results, as well as preliminary work on the evaluation of secondary metabolites produced by mycelial cultures of *Laccaria* spp., will be presented.

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PS - poster session

**Combinatorial impact of different shocks on the composition of membrane lipids and osmolytes in *Aspergillus niger***

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Under natural conditions organism is often exposed to the simultaneous action of several stress factors, but such combinatorial stresses in fungi are not sufficiently studied. The key elements of the defense system are changes of the membrane lipid composition and osmolytes. Our previous comparative study of responses to individual shocks in *Aspergillus niger* made it possible to continue with the present research: the impact of different combinations of osmotic (OS), heat (HS) and oxidative (OxS) shocks on the composition of soluble cytosol carbohydrates and membrane lipids.

For the first time it was demonstrated that the combinatorial effect of HS and OS led to the non-additive response - an increase in the trehalose level, characteristic for HS, but, at the same time, suppression of glycerol production, uncharacteristic for the OS response. In addition, new effect was observed - increase in the mannitol level, which was not typical for the individual HS or OS responses.

OxS, caused by higher concentration of H<sub>2</sub>O<sub>2</sub> (50 mM), in combinations with HS or OS led to the decrease of the total amount of carbohydrates, mainly due to the drop of the mannitol level. At the same time, OxS pairwise combined with other shocks, inhibited trehalose or glycerol synthesis, characteristic for individual HS or OS responses respectively.

Thus, responses to different combinatorial shocks include changes of carbohydrate composition, characteristic for individual shocks, as well as new effects. On the contrary, general pattern of change was observed in the composition of membrane lipids in response to all shock combinations: an increase in the proportion of phosphatidic acids.

The study was supported by the Russian Foundation for Basic Research (project 18-34-00230).



PS - poster session

**Genetic determinants and cellular processes involved in the interactions of ectomycorrhizal fungi with toxic metal species**

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Mycorrhizal fungi play an important dual role in plant metal homeostasis: scavenging of metal micronutrients and their supply to the host; detoxification of both the excess essential and physiologically irrelevant metals. This report focuses on the handling of metals in the species accumulating remarkably high concentrations of heavy metals. Searches of transcriptomes for metal related determinants, gene complementation studies in yeasts, metal speciation analyses in fungal tissues, and gene expression analyses allowed us to obtain the evidences supporting the following conclusions. In Ag-hyperaccumulating *Amanita strobiliformis*, at least two CTR transporters can efficiently recognize for import Ag in addition to Cu, P<sub>1B-1</sub>-ATPase transporter can export Ag and Cu from the cytoplasm but virtually all the accumulated Ag occurs complexed by cytosolic metallothioneins (MTs). In Zn over-accumulating *Russula bresadolae*, which has a high affinity ZIP transporter for the acquisition of Zn and CDF transporter for vacuolar sequestration of overaccumulated Zn, is 40% of accumulated Zn bound with unusual MT-like RaZBP peptides; RaZBP homologues are involved in binding of the excess of intracellular Zn also in *Russula* spp., which form with *R. bresadolae* a separate phylogenetic subclade of Zn overaccumulating species. In contrast, *Hebeloma mesophaeum*, like several other *Hebeloma* species, preferentially funnels excess Zn (and Cd) into subcellular compartments, although it has the capacity to produce MTs. Altogether, our data suggest that there might be a link between cellular biology of Ag and Cu in ectomycorrhizal fungi and show that fungi of different genera (*Russula* vs. *Hebeloma*) may employ different strategy to handle the excess of particular metal (Zn).

PS - poster session

**The importance of inoculum volume for the competitive outcome and wood decay ability of brown- and white-rot basidiomycetes**

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Theory predicts that energetic cost for competition between fungal mycelium might accelerate or retard the rate of wood decomposition, depending on various factors. To evaluate the effect of inoculum size on competitive outcome and wood decay rate, we set up pairing competition experiment using 8 cm<sup>3</sup> beech wood blocks colonised by each of 3 brown-rot (*Coniophora puteana*, *Laetiporus sulphureus*, *Fomitopsis pinicola*) and 3 white-rot basidiomycetes (*Hypholoma fasciculare*, *Trametes versicolor*, *Vuilleminia commedens*). Four wood blocks were used in each competition, and four levels of inoculum volume were established for each fungus: (1) one block was inoculated with species A and three blocks with species B; (2) both of the species with two blocks each; (3) three blocks with species A and one block with species B; (4) all of the four blocks with species A (control). Paired blocks were incubated in the dark at 20°C for 3 months, and then competition outcome and wood density loss were recorded after the harvest. Results showed that larger mycelium outcompeted smaller mycelium. Paired competition generally accelerated but occasionally reduced wood density loss. Effect of inoculum wood volume on decay rate during competition was fungal species dependent (e.g. negative for *Hypholoma* but positive for *Fomitopsis*), and we could not find any clear difference between brown- and white-rot fungi. In some cases, wood density loss was slower in the competition front than in the rear of the wood block. These results suggest that competition itself may accelerate wood decay, but the effect varies depending on the location within the mycelium, and the effect of inoculum volume was fungal species dependent.

PS - poster session

**Bacterial endosymbionts of Mucoromycota representatives**

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Fungal-bacterial interactions is a relatively new area of research which challenges our perception of plant terrestrialisation. Presence of endosymbionts in fungi from Mucoromycota phylum (which are considered to be the most ancient group of plant-related fungi and to have helped plants in land colonisation<sup>1</sup>) supports the hypothesis that the first land plant was rather a consortium of algal, fungal, and bacterial cells than an algal colony.

First clues that some fungi possess “bacteria-like structures” were found nearly 40 years ago but actual evidence of intimate relationship between fungi and bacteria dates back to 2003<sup>2</sup>. Although there are only a few described partnerships between mucoralean fungi and bacteria, like *Gigaspora margarita* and *Ca. Glomeribacter gigasporarum*<sup>2</sup>, *Rhizopus microsporus* and *Paraburkholderia* spp.<sup>3</sup>, or *Mortierella elongata* and *Mycoavidus cysteinexigens*<sup>4</sup>, it is still not known what is the purpose of these symbioses or even how widespread are they across Mucoromycota phylum.

In order to check how often mucoralean fungi harbour endohyphal bacteria (EHB), we performed a PCR-based screening of Mucoromycota representatives for the presence of EHB. Our results suggest that ca. 15% of *Mortierella* strains harbour EHB belonging to Burkholderiaceae family (BRE) and this finding is consistent with studies conducted previously<sup>4,5</sup>. We also report BRE from the hyphae of *Umbelopsis*. Among bacteria we detected, there are four lineages of BRE which were not described before.

Following step would be genomic analysis of the holobionts with aim of finding the genes responsible for establishing relationship between the partners. Long-term goal of the study is understanding the nature of these symbioses and assessing their possible role in land colonisation.



PS - poster session

**The impact of fourteen tree species on arbuscular mycorrhizal fungi abundance, species richness and composition in soils of the Siemianice Experimental Forest**

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The overwhelming part of mycological studies in forests of temperate climate zone focuses on ectomycorrhizas, nevertheless also endomycorrhizas, especially arbuscular mycorrhiza (AM), among forest understorey and overstorey species have been observed. Despite the presence of arbuscular mycorrhizal fungi (AMF) in such ecosystems, there is still insufficient knowledge on their diversity and interactions with plants. Therefore, we studied the impact of overstorey specificity on AMF abundance, species richness and composition, in relation to herbaceous plant cover and soil chemical properties. The effects of 14 tree species grown in monospecific plots for 48 years in the Siemianice Experimental Forest (central Poland), including groups of deciduous vs. coniferous, native vs. alien to Europe, forming vs. not forming AM, were compared. AMF spore and species numbers were at low level in comparison to other unforested ecosystems, and averaged 1.6 and 0.02 per 1 g of soils, respectively. The presence of 8 AMF species, both widespread (e.g. *Funneliformis constrictus*) and rare (i.e. *Acaulospora cavernata*) was revealed. There was significant divergence between AMF species composition in plots of deciduous and coniferous species. The concentrations of PLFA 16:1 $\omega$ 5 AMF hyphal biomass marker were higher in soils of deciduous and AM-type tree species than coniferous and non-AM ones, also the concentrations of NLFA 16:1 $\omega$ 5 AMF spore biomass marker were higher in soils of deciduous tree species. No significant differences between native and alien to Europe groups of trees were found. AMF abundance in soil and roots increased along with increasing soil alkalinity and macronutrient levels.

Financing: National Science Center, Poland (2017/27/N/NZ8/00999).



PS - poster session

**Rhizosphere and endophyte microcommunities of *Trifolium repens* from Zn-Pb waste heap (S. Poland)**

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Waste-heaps created as the deposits of post-mining zinc and lead ores are highly heavy metal polluted areas, additionally deficient in water and nutrients. Heavy metals present in such industrially modified soil may act as a factor of the natural selection pressure, shaping the taxonomical structure of the microcommunities. As the result, the genotypes of the high resistance to heavy metals may be fixed, which usually are able to improve the growth of plants, and those with a remediative potential too. Automated ribosomal intergenic spacer analysis is the one of metagenome tools, culture-independent, PCR-based and automated technique used for detection of taxonomical genetic diversity of environmental microcommunities, based on the capillary electrophoresis of 16S-23S rRNA fragments. The aim of present study was to estimate the metagenome polymorphism of fungal and bacterial rhizosphere and endophyte (roots, nodules and leaves) communities of white clover (*Trifolium repens*), inhabiting the old, about 100-yr-old Zn-Pb Bolesław waste heap (S. Poland), with compare to control microcommunities according the ARISA course.



PS - poster session

**Ectomycorrhizal community composition changes along to silver fir (*Abies alba* Mill.) phenological stages in Slovenia**

Tina Unuk Nahberger, Rok Damjanić, Hojka Kraigher, Tine Grebenc  
*Slovenian Forestry Institute, Ljubljana, Slovenia*

In forest ecosystems, tree roots are colonized by various ectomycorrhizal fungi, which impact plant growth and vigour, as are recognized as potential drivers of nutrient mobilization processes of their host plants. As we in study Unuk et al. (2019) observed statistically significant differences in ectomycorrhizal community between phenologically distinct groups of silver fir trees, our aim in this study was to analyze ectomycorrhizal fungal community of silver fir and changes in abundance linked to phenology. The phenological stages of silver fir trees were followed weekly for two years, and along these, 5 soil cores for each observed silver fir tree were analyzed for ectomycorrhizal symbionts, which were further identified using Sanger sequencing of an individual ectomycorrhizal morphotype. Analyzes of beta diversity between phenological stages revealed significant differences in the overall ectomycorrhizal community between dormancy and bud bursting, whereas there is an overlap between ectomycorrhizal community in dormancy and full needle expansion as well as between bud bursting and full needle expansion.

Abundance analyzes of ectomycorrhizal species between phenological stages revealed higher abundances of *Russula ochroleuca*, *Tomentella stuposa*, *Tomentella sublilacina* and *Tylospora fibrillosa* during bud bursting, which are ectomycorrhizal species for which the presence of manganese peroxidase and/or laccase has already been suggested or confirmed. Our results indicate a need for more detailed analyzes of possible presence of mentioned enzymes among ectomycorrhizal species associated with bud burst and silver fir trees.

The presentation contributes to the aims and means of the project LIFE GENMON (LIFE13 ENV/SI/000148).



PS - poster session

**Biological control potential of ectomycorrhizal fungi against *F. circinatum* on *Pinus patula* seedlings**

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Ectomycorrhizal fungal relationships are well known to act biological control agents for their hosts along with providing increased access to nutrients and water. In the forestry industry, the fungal pathogen *Fusarium circinatum* is one of the largest limitations to commercial forestry both on plantations and within nurseries. In the present study, the biological control and growth promoting ability of local ECM fungi as an inoculum for *Pinus patula* seedlings exposed to *F. circinatum* strains is investigated. Four ECM fungal strains were isolated and molecularly identified as *Boletus edulis*, *Lactarius quieticolor*, *Suillus granulatus* and a *Suillus* strain. Their anti-fungal activity was shown to be highly varied in a dual assay with six *F. circinatum* strains: FC666, FC594, FC621, FC623, and FC701. The combined results of the anti-fungal assay and greenhouse trial showed that *B. edulis* and *S. granulatus* both produced indirect forms of pathogen inhibition while the *Suillus* strain and *L. quieticolor* both had much more direct forms of inhibition. Overall inoculation the *Suillus* strain and *L. quieticolor* resulted in the most significant growth of the *P. patula* seedlings. Thus, it is these isolates which require further research in the hopes of developing a commercial ECM inoculum for the South African forestry seedling industry to help combat *F. circinatum* and the generally improve the health of *P. patula* seedlings in the nursery and field.



PS - poster session

**Do dark septate endophytes parasitize plant parasitic nematodes?**

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Dark septate endophytes (DSEs) are globally distributed root-colonizing endophytic fungi and found in a great variety of host plants. They are especially frequent in harsh environments. Nevertheless, their function in plants is still elusive. Cereal cyst nematodes (CCNs) are obligate parasites and establish a long-term biotrophic interaction with the host plants. The impact of CCNs can be especially severe in semi-arid regions where abiotic stresses often occur. Observations on cysts of the CCN *Heterodera filipjevi* from wheat fields in Turkey revealed the presence of many nematode eggs colonised by fungi causing different symptoms. A study was therefore conducted to characterize these symptom-causing fungi, study their interaction with *H. filipjevi*, and profile their secondary metabolites. Three novel ascomycetous species were isolated from infested nematode eggs. Phylogenetic analyses revealed that they belong to the Helotiales and Pleosporales. The sequences obtained from pure cultures were identical to those obtained from individual nematode eggs confirming identity of the symptom-causing fungi with the isolated pure cultures. Koch's postulates were fulfilled confirming that all three isolates were able to infect healthy eggs in vitro. All three species were found to be root endophytes in independent studies. The newly described helotialean DSE species *Polyphilus sieberi* was also isolated from different plant species. Phylogenetic analyses suggest that the two newly found pleosporalean species are also conspecific to some reported root endophytes. Four new cyclodepsipeptides isolated from one of the pleosporalean strains showed nematicidal activity against *H. filipjevi* in vitro. To conclude nematode eggs seem to represent a niche where unknown endophytes can be found

PS - poster session

**Stem rust infection in Novosibirsk region: life cycle and origin**

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Stem rust, caused by *Puccinia graminis* f. sp. *tritici* (Pgt) has lately gained importance for Siberian wheat crops. All stages of the full life cycle of a pathogenic fungus have been found in the Novosibirsk region (sexual reproduction on the intermediate host, barberry, and asexual reproduction on wheat and grasses — urediniospores and teliospores). However, it is unknown whether all the detected infection patterns belong to the same special form of the fungus. The objective of this study was to examine the life cycle of Pgt in the Novosibirsk region and to establish the origin of the local pathogen population.

Samples of stem rust were collected in 2016-2017 from leaves of barberry, wheat stems and grasses in the Novosibirsk region.

Field and laboratory virulence tests have shown that the barberry is not a source of infection for wheat crops in the Novosibirsk region. Phylogenetic analysis with the involvement of 63 ITS sequences of stem rust from GenBank showed that the fungus sequences isolated from the leaves of the barberry are clustered with the forms specific for grasses. Most of tested SSR-markers (9 of 11) gave the same profiles for samples of aeciospores and urediniopores collected from wild grasses, but not from wheat.

Full life cycle in the Novosibirsk region for a special form of the fungus that cannot infect wheat is realized. The population of *P. graminis* on wheat is airborne for the Novosibirsk region.

This work was supported by the Russian Foundation for Basic Research № 17-29-08018 and budget project № 0259-2019-0001.

PS - poster session

### **How do mycorrhizal fungi cope with metal stress?**

**Olga Bogdanova**, Anna Bonrath, Katrin Krause, Erika Kothe  
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Microorganism assisted phytoremediation is an efficient approach for rehabilitation of heavy metal polluted soils. Here, we focus on ectomycorrhizosphere as an area of close interaction between plant roots, ectomycorrhizal fungi, microorganisms and soil. The effect of bacteria isolated from ectomycorrhizosphere of the plants naturally growing at heavy metal contaminated site on the growth of ectomycorrhizal fungi is assessed. The growth of the fungus *Tricholoma vaccinum* in co-cultures with *Streptomyces coelicolor* (metal-sensitive) or *Streptomyces mirabilis* (metal-resistant) in the presence of nickel was estimated. The growth of *T. vaccinum* was induced in divided plates in co-cultures with *S. coelicolor* (interaction by volatiles) whereas in co-cultures with *S. mirabilis* increased growth was observed in regular plates (direct influence of metabolites excreted by the bacterium).

In this study we focused also on hydrophobins, which are unique surface-active fungal proteins. Up-regulation of *hyd8* gene during the formation of aerial mycelium by *T. vaccinum* as well as under the metal stress was shown earlier; however, direct participation of hydrophobins in the alleviation of metal stress has not been addressed so far (Sammer et al., 2016). In our experiment the rodlet surface of two transformants overexpressing *hyd8* in the presence of nickel and copper was investigated with transmission electron microscope. It was determined that metal treatment resulted in changes in rodlet surface of transformants, for example, elongation and number of rodlets per package. Further experiments to assess the role of *hyd8* in mycorrhiza establishment under the metal stress will be performed.

Both studies will help to understand the mechanisms involved in metal stress in the ectomycorrhizosphere.



PS - poster session

**Molecular, physiological and anatomical changes in common wheat (*Triticum aestivum* L.) seedlings during interaction with *Trichoderma* fungi**

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Fungi of the genus *Trichoderma* (teleomorph *Hypocrea*) are free-living organisms and are widespread in the environment. They occur in all climate zones and colonize different ecological niches. These fungi are producers of many secondary metabolites which play a key role in their interaction with plants and other microorganisms. In this study we use two species of *Trichoderma* differentiated in terms of lifestyle and production of metabolites: *Trichoderma atroviride* strain AN35 - an efficient producer of glucanases and volatile metabolites, including 6-npentyl-2H-pyran-2-one, with the highest antagonistic potential towards mycotoxigenic *Fusarium* spp. and *Trichoderma cremeum* strain AN392 – representing saprotrophic lifestyle an efficient producer of cellulolytic and xylanolytic enzymes. Both of *Trichoderma* species originated from Poland, with that *T. cremeum* AN392 was obtained from sample of decaying wood collected in the forest of the Gorce Mountains located in Poland and *T. atroviride* AN35 was isolated from maize kernels.

The main goal of this study was to analysis of the changes in the morphology, anatomy, physiology and selected gene expression in wheat seedlings after *Trichoderma* strain inoculation. Our results show that *Trichoderma* spp. can colonize roots of wheat seedlings and have impact on wheat plants growth component, yield parameters, chlorophyll fluorescence, roots length and thickness of roots and leaves, and also on plant hormones (like auxins, zeatin, kinetin, salicylic acid, gibberellin, jasmonic acid, abscisic acid) production in roots, and on the expression of selected genes known to be related to defence and resistance in wheat.

This work is supported by National Science Centre (NCN, Poland), research project No. 2015/19/B/NZ9/03083.



PS - poster session

**Deciphering casing microbiome: effect of fungal infection and pesticide treatment in button mushroom crops**

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The cultivation of edible mushroom is an strategic sector for the economy of La Rioja, the most productive region of Spain, that requires intensive hand labour, and therefore contributes to fix population in low density rural areas. It intrinsically consists of the development of selective substrates through composting where the mushroom grows via solid fermentation process. The compost fully colonized by the *Agaricus* mycelium hardly produces mushrooms and it is necessary to apply a casing layer which requires certain physical, chemical and biological characteristics to reproduce the appropriate environment in which takes place the morphological shift from the vegetative mycelium to the reproductive one, where the native microbiota plays crucial roles. Currently, the industry faces a challenge to substitute the actual peat based casing materials due to the limited natural resources and the impact on the peatlands where peat is extracted.

In this work we have employed high-throughput techniques by next generation sequencing to screen the microbial structure of different casing materials employed in mushroom cultivation while sequencing V3-V4 of the 16S rRNA gene for bacteria and the ITS2 region of rRNA for fungi in an Illumina MiSeq. In addition, the microbiome dynamics and evolution (bacterial and fungal communities) in peat based casing along the process of cultivation of *Agaricus bisporus* have been studied, while comparing the effect of fungicidal treatment (chlorothalonil and metrafenone), and the impact of artificial infection with fungal parasites (*Lecanicillium fungicola* and *Mycogone perniciosa*).

Project funded H2020 programme under the Marie Skłodowska-Curie grant agreement no. 742966 and also funded by the ADER project grant no. 2016-IDD-00013.



PS - poster session

**Identification and characterization of a new wasting disease affecting commercial exploitations of *Pleurotus ostreatus***

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The commercial cultivation of edible mushrooms is one of most important agricultural economic sectors in La Rioja. In the last few years, the growths of the oyster mushroom (*Pleurotus ostreatus*) have been seriously affected by a wasting disease of unknown origin, responsible for important economic losses. This study was designed to better understand the morphological changes experienced by the carpophores.

Several specimens of *P. ostreatus* were collected in local farms. Some of them were healthy whereas others were affected by the disease. Diseased mushrooms had a compromised hyphal architecture, losing in some cases the external palisade, allowing some bacteria to colonize the mushroom body. This only happened in the most affected specimens, but most diseased mushrooms did not have any bacteria inside their fruiting bodies, thus indicating that the disease is most likely not caused by a bacterial infection. Interestingly, diseased hyphae were more basophilic than the healthy ones, suggesting a metabolic acidification due to the disease. Semithin sections provided additional information on the morphology of the diseased hyphae that were characterized by a darker and denser cytoplasm. The number of affected hyphae correlated with the severity of the disease. Electron microscopy showed that diseased hyphae presented a retracted cytoplasm, a large thickening of the cell wall, and a number of small vesicles in the cytoplasm (30-60 nm in diameter) consistent with a viral infection.

Morphological characterization of the diseased specimens of *P. ostreatus* suggests that the wasting disease responsible for important economic losses in commercial growths is not due to the presence of viral particles of unknown origin.

Project funded by the ADER project grant no. 2018-IDD-00050.



PS - poster session

**Characterization of an extracellular laccases and decolorization of synthetic dyes from mycelium of *Lentinula edodes* (shiitake)**

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*Lentinula edodes* (Shiitake) is an important edible mushroom in the world. *L. edodes* is distributed in Asia and some subtropical regions and is a white rot fungus with excellent wood-degrading ability. The lignin activity of this species can be measured using laccases, which are multi-copper oxidase that catalyze the transformation of aromatic and non-aromatic compounds with reduction of molecular oxygen to water. Synthetic dyes are phenolic compounds which can be used to measure laccase activity. In this study, we investigated the correlation between the growth of strains and laccase activity by using the decolorization of different synthetic dyes and measure vitality according to the storage period. Mycelial growth, chromaticity change, dye decolorization and laccase activity were measured using six synthetic dyes for ten strains of this species. In the color of media containing methyl red, mycelial growth was slow and chromaticity change was weak. However, in the color of media containing bromocresol purple, bromothymol blue and congo red, the chromaticity change shows high and has higher correlation than other dyes. The correlation coefficient among dye decolorization, mycelial growth, and laccase activity was not so significant. In the color media using bromothymol blue, decolorization ratio and laccase activity showed higher correlation than other dyes. In preserved strains, the longer stored media showed the less mycelial growths and a low laccase activities. These data suggest that these processes are considered to be an useful methods for measuring the viability of fungal strains.



PS - poster session

**Spread of *Heterobasidion* root rot infection in Norway spruce stands on former agricultural land after thinning**

**Darta Klavina**, Lauma Bruna, Natalija Burnevica, Talis Gaitnieks  
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Reforestation of former agricultural lands is an important issue in Europe and Norway spruce (*Picea abies* (L.) H.Karst.) is one of the most frequently used tree species for this purpose. Such spruce stands are characterised by high wood productivity but also - high infection risk of root rot fungi (*Heterobasidion* spp.) after thinning.

To detect whether *Heterobasidion* root rot is present in spruce stands on previous agricultural lands after thinning we surveyed 13 spruce stands in Latvia and collected wood samples of 3 to 10 trees if some disease symptoms were observed. Within these stands, five had no infection, four had low disease incidence while four stands were heavily infected. From those we choose three stands for analysis of genets of *Heterobasidion* spp. and its spread among trees. Our aim was to determine spread of root rot infection among the trees via root contacts. Five to seven circle sample plots (10m in a radius) per stand were established and wood samples were taken from all standing trees and stumps. Under lab conditions, presence of *Heterobasidion* spp. mycelium and conidia in those was detected and fungal isolates obtained. By somatic incompatibility, information about number of genets and its size was obtained. Results present spread of *Heterobasidion* root rot in those stands within period from thinning.

In further work, assessment of economic losses caused by *Heterobasidion* root rot on former agricultural lands and analysis of fungal communities in roots and wood is planned.



PS - poster session

**Ecophysiology and virulence of *Pseudogymnoascus destructans***

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The fungus *Pseudogymnoascus destructans* (Ascomycota) invades the skin of hibernating bats. This pathogen was introduced to the US, infecting naive hosts and causing “white nose syndrome”, resulting in bat mortality. Our multidisciplinary team has a long time interest in the complex understanding of *P. destructans* biology. We discovered the cryptic sexuality in the Czech populations and our sampling from Ural expanded its source region. We compared the metabolome of virulent and non-virulent *Pseudogymnoascus* species and identified an alkalization ability, a lipase activity, siderophore production and hyperproduction of riboflavin as features specifically linked with *P. destructans*. Riboflavin itself has distinctive yellow-orange fluorescence, but its possible hyperaccumulation in the skin lesion needs a further study. The properties related with unique ecology of *P. destructans* were studied by comparisons of tolerance to environmental stresses and spectra of utilized nutrients across various ecologically diverse species. *P. destructans* didn't differ from other cave fungi by the stress tolerance, but has a unique physiological profile, typical for specialists, setting it apart from cave saprobic and dermatophytic fungi.

PS - poster session

**Effects of season and bedrock on ectomycorrhizal community composition in  
Submediterranean oak secondary forest**

**Tanja Mrak**, Nataša Šibanc, Ines Štraus, Jožica Gričar, Hojka Kraigher  
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Slovenian Submediterranean is characterized by two types of bedrock, flysch and carbonate. Soils on carbonate were eroded to a large degree in the past due to human activities, forming a typical stony landscape, Karst. After abandonment of pastures, area was spontaneously encroached by various woody species with dominant pubescent oak (*Quercus pubescens* Willd.). Pubescent oak is ecologically very important for this area, as it prevents further degradation of shallow soils. Harsh abiotic factors (frequent drought and wildfires) are strongly limiting plant growth in this area, but on the other hand, mycorrhizal fungi may have beneficial effects. To characterize ectomycorrhizal fungal (EMF) communities of pubescent oak on two types of bedrock, soil cores from calcareous and flysch plots were collected from June 2016 till May 2018. From each soil core, ectomycorrhizal morphotypes associated with oak roots were isolated, morpho-anatomically characterized, and sequenced using ITS1/ITS4 markers. Assembled sequences were identified using UNITE database and molecular identification was compared and combined with morpho-anatomical identification. Community composition of EMF was affected by season and bedrock. Calcareous plot was characterized by higher species richness and lower seasonal variability of species richness compared to flysch plot. Maximum of species richness occurred at both types of bedrock at the same time. For the whole period and both plots, around 70 morphotypes of EMF were listed. Significantly higher abundance of *Cenococcum geophilum* (Gloniaceae, Ascomycota) was recorded for calcareous plot, and *Sebacina* sp. (Sebacinaceae, Basidiomycota) for flysch plot. The lowest percentage of vital ectomycorrhizal tips was detected in 2017, a year following wildfire.



PS - poster session

**Isolation and characterisation of metallothioneins from Cd-accumulating *Cystoderma carcharias***

**Tereza Leonhardt**<sup>1</sup>, Jan Sácký<sup>1</sup>, Jiří Černý<sup>1</sup>, Jan Borovička<sup>2</sup>, Pavel Kotrba<sup>1</sup>

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The natural ability of fungi to accumulate significant amounts of various heavy metals in their sporocarps and mycelia has been known for decades. While for most macroscopic fungi, Cd concentrations were reported in units to tens of mg Cd kg<sup>-1</sup> of dry weight, in the saprotrophic fungus *Cystoderma carcharias* measured concentrations were up to 600 mg of Cd kg<sup>-1</sup> of dry weight, which is the highest level of Cd accumulation for fungi published so far. Besides Cd, *C. carcharias* is an efficient accumulator of Ag, Cu and Zn. In this work we have isolated two metallothioneins, CcMT1 and CcMT2, and assessed their ability to protect metal-sensitive yeast mutants from Cd and Zn poisoning. The MTs both contain Cys-AA-Cys motifs typical of metallothionein sequences and share nearly 73 % similarity, but the differences in the sequences resulted in CcMT1 being more efficient for Cd and Zn detoxification, while CcMT2 seems to be more efficient for Cu detoxification in yeast mutants.

PS - poster session

**Novel aluminium-binding compound from *Oidiodendron* sp.**

**Svetlana Baiandina**, Tereza Leonhardt, Antonín Kaňa  
*UCT Prague, Prague, Czech Republic*

Ericoid mycorrhizal fungi are associated with plants from the order Ericaceae, some of which are of nutritional value to humans, such as cranberry or blueberry. The mycorrhizal symbiosis is beneficial to the plant also with regards to decreased heavy metal accumulation when grown on metal polluted soil. Several metal-tolerant *Oidiodendron* species have been already reported, but only very few genetic determinants related to heavy metal metabolism have been characterised. A metal tolerant *Oidiodendron* isolate has been propagated in laboratory settings with high levels of various heavy metals (Al, Cd, Cu, Ni, Zn) and speciation of these metals in the mycelial biomass has been resolved by size exclusion chromatography with mass spectrometry detection. Novel Al-binding 2-kDa compound has been detected in the mycelium of *Oidiodendron* sp. This Al-binding compound will be investigated further to discern its origin.

PS - poster session

**Arsenic accumulation and speciation in *Russula pumila***

**Gabriela Pelešková**<sup>1</sup>, Jan Sáčký<sup>1</sup>, Jan Borovička<sup>2</sup>, Simone Braeuer<sup>3</sup>, Walter Goessler<sup>3</sup>, Pavel Kotrba<sup>1</sup>

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Arsenic (As) is a toxic metalloid with no known biogenic function. To prevent its toxicity, organisms employ a variety of metabolic processes to transform arsenate (As V) that they obtain from the environment into less toxic compounds. These compounds are then stored within the cell in specialised compartments or exported out of the cell. *Russula pumila*, an ectomycorrhizal fungus growing on marshes, preferably with alder trees, has been reported as an efficient As accumulator (up to 400 mg As kg<sup>-1</sup> dry weight). However, the speciation and mechanism of As accumulation and storage are not yet known. Interestingly, *Russula pumila* is also a Zn accumulator and from size exclusion chromatography of sporocarp extracts it would seem that the As and Zn species are both of peptidaceous nature. However, none of the Zn-binding peptides confer resistance to As in As-sensitive yeasts mutants. In this work we try to elucidate the speciation of As in *R. pumila* sporocarps by size exclusion chromatography and ion exchange chromatography with mass spectrometry detection of As species.



PS - poster session

**Fungal microbiota of caterpillars and phylloplane – how much do they differ?**

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Mycobiota of phylloplane are considered as one of the most diverse microbial assemblages. Conversely, the microbiota of the caterpillar gut are assessed as a poor due to strongly alkaline gut conditions. So far, in herbivorous insects, more attention has been paid to bacterial microbiota, whereas fungal ones were neglected.

We aimed to 1) Compare richness and betadiversity of the gut and phylloplane fungal microbiotas; 2) Quantify their overlap (similarity); 3) Identify MOTUs significantly associated with them. Fungal microbiota was identified by DNA metabarcoding (gene ITS) and data were analysed using rarefactions, GLMs, Multi-level pattern analysis and partial RDA.

Fungal assemblages were richer than bacterial in majority of samples for phylloplanes and in half of samples for caterpillar gut. Betadiversity of fungal assemblages was higher in caterpillar gut than in phylloplane and their similarity was quite low (38.12%), suggesting that caterpillar gut harbour separate fungal microbiota, as was confirmed also by RDA. Fungal MOTUs most significantly associated with the caterpillar gut were *Ophiostoma*, *Mucor* or *Cyberlindnera*, whereas MOTUs associated with phylloplane were *Holtermanniella*, *Gibberella*, *Seimatosporium* or *Mycoleptodiscus*. The results suggest that the fungal microbiota of the caterpillar gut is not entirely derived from phylloplane and, therefore, it represents an attractive environment for further research taking into account its specific diversity.

Research was funded by the Ministry of Education of the Czech Republic (LO1208), Inst. of Env. Technologies (CZ.1.05/2.1.00/03.0100, (CZ.1.05/2.1.00/19.0388), NextGenProject (CZ.1.07/2.3./20.0303), University of Ostrava (SGS23/PřF/2016, SGS15/PřF/2017 & SGS14/PřF/2018) and Czech Science Foundation (GA18-08803S).





PS - poster session

**Aclinic method of trap culture for arbuscular mycorrhizal fungi**

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Trap culture of arbuscular mycorrhizal fungi (AMF) on plant cuttings was studied in small plastic boxes and cultivated in horizontal position. Such system was tested for mycorrhization of Lamiaceae on cultivars of model plant *Plectranthus scutellarioides*. Boxes of the same size (95x85x20 mm) with a cover which had 60x5 mm hole in a side wall were exploited for cultivation. Tested cuttings of *P. scutellarioides* had been rooted in sterile water before inoculation with AMF. Single rooted cutting was placed into the box, later filled in with steam sterilized quartz sand of 1.4–2.0 mm fraction and humidified with 20 ml of sterilized water. Spores, isolated by wet sieving and sucrose gradient centrifugation, and root fragments of mycorrhized plants from the collection of standard trap culture in pots were used as AMF inoculum. Processed boxes with inoculated cuttings were incubated horizontally in piles inside grow chamber at 22-24° C, 16 h light period and 17-19° C, 8 h dark period. The aclinic system occupies less space and is more effectively controlled in comparison to conventional pot cultures. Application of *P. scutellarioides* rooted cuttings in tested aclinic method prevents some potential contamination of the system by pathogenic and saprotrophic fungi that was confirmed during monitoring. Preliminary results of sterilized seed germination of *Sorghum bicolor* under the same conditions of the aclinic system had demonstrated high risk of other fungi growth. AMF lifecycle stages were easily studied under the dissecting microscope as a part of plant root system grows on surface and in upper layer of quartz sand due to uniform water balance inside the box. Water condensation on the cover inner surface facilitated AMF development in the upper layer of quartz sand.

PS - poster session

**Can species belonging to *Mortierella lignicola* clade be associated with temperate forest ant species?**

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*Mortierella* is a fungal genus common in soil, used in biotechnology as a valuable source of polyunsaturated fatty acids<sup>1</sup>. Interestingly, species belonging to *Mortierella lignicola* clade are known not only as soil saprotrophs but are also often isolated from ants<sup>2,3,4</sup>. In the study of Clark (2002), strains of *M. beljakovae* were isolated from infrabuccal pockets of *Camponotus pennsylvanicus* and *C. herculeaneus* ants. In our studies, we have isolated *M. beljakovae* and *M. lignicola* from the bodies of *F. polyctena* ants, *M. beljakovae* from *F. pratensis* and *M. gemmifera* from *F. cinerea* ants. Moreover, from *F. pratensis*, we have isolated a new species: *M. formicae*. One of the characteristics of species belonging to *M. lignicola* clade is the ability to form numerous gemmae filled with oil droplets. However, the unique characteristics of *M. formicae* is the formation of dense and thick clusters of gemmae. Importantly, reproductive form (asexual) of this species was obtained only in stressful conditions, one year after inoculation. In the fatty acids profile analysis of *M. alpina* and *M. formicae* we discovered that the mycelium of *M. formicae* contains significantly more saturated fatty acids than the mycelium of *M. alpina*. Interestingly, in insects, fatty acids play a significant role in the production of pheromones and defensive secretions<sup>5</sup>. In the behavioral tests, in which we presented *M. formicae* to *Formica* s. str. ants, the ants were highly interested in the fungi, often performing licking behavior and sometimes even transporting hyphae to the nest. The preliminary results of our studies show that the species belonging to *M. lignicola* clade are associated with ants. Further studies are needed to answer if the specific biology of *M. formicae* are a result of ant-fungal co-evolution.



PS - poster session

**Metagenomic analysis of rhizosphere soil around *Amanita caesarea*, *A. verna*, *Cortinarius violaceus*, *Laccaria vinaceoavellanea* in Jeju island in Korea**

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Gothawal in Jeju Island, a volcanic island of lava layers in Korea, is considered an internationally important wetland which is within the temperate mixed forest biome. To study ecological influence of rhizosphere soil on *Amanita caesarea*, *A. verna*, *Cortinarius violaceus*, *Laccaria vinaceoavellanea* mushrooms, microbial communities were analyzed from these four mushrooms rhizosphere soils using the microbiome taxonomic profile(MTP). Operational taxonomic units(OTUs) which is based on internal transcribed spacer(ITS) for fungus and 16s rRNA V3V4 region for bacteria were used for the analysis. The most diversity of OTUs was found in rhizosphere soil of *A. verna* followed by that of *L. vinaceoavellanea*, *A. caesarea* and *C. violaceus*, respectively. Species richness was considerably higher in the rhizosphere soils of *A. verna* and *L. vinaceoavellanea* than in those of others. Regarding MTP taxonomic composition at the level of bacterial phylum, the most abundant bacterial phylum was Proteobacteria, followed by Acidbacteria, while Bacteroidetes was dominant in the rhizosphere soil of *A. verna*. The most abundant phylum of fungi in the four rhizosphere soils was Basidiomycota, followed by Ascomycota and Mortierellomycota. Each rhizosphere soil showed a slightly different pattern for each soil. This finding suggests that the difference in the abundance of the fungi and bacteria in the rhizospheres soil of mushroom from Jeju Island may have been affected by geographic location and special climate. Our study is the first detailed report of the composition of soil fungal and bacterial communities in mushroom rhizosphere soil from Jeju island. In addition, our findings provide a basis for understanding the ecological interactions among the mushroom species and soil fungi and bacteria.



PS - poster session

***Puccinia brachypodii* a rare rust fungus with low genetic diversity**

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*Puccinia brachypodii* G.H. Otth (syn. *P. arrhenatheri* (Kleb.) Erikss) is a pathogen of *Berberis* genus, causing witches' brooms on them. Leaves on witches' brooms appear earlier than those on the uninfected part of the bush and are light coloured, overgrown by rust mycelium. *P. arrhenatheri* is considered as conspecific with *P. brachypodii*. *Arrhenatherum elatius*, *Melica nutans* and other *Poace*, including *Apera spica-venti* can serve as the second host of it. *P. brachypodii* is a worldwide (Europe, Asia, North part of Africa) and it seems to be more destructive for barberry bushes than for grasses, causing their slow dieback.

The life cycle of rust is considered as macrocyclic, what means that it proceeds through five possible spore stages on two different host plants. The sexual reproduction goes on *B. vulgaris* – to complete dicarotic phase, for this purpose the fungus forms spermogonia. Aecia are usually formed on the bottom side of infected leaves. Lack of compatible mating types of mycelia, in the same environment, leads to incomplete life of *P. brachypodii* with no aecial phase. Systemic mycelium overwinters within barberry bushes. *P. arrhenatheri* uses scent attraction for insect mediated exchange of mating-type gametes (spermatia), for this purpose the bright yellow colour of mycelia on leaves is created. Volatile compounds used as attractants for insects include chemicals which are responsible for distinct floral-fruity and herbaceous scent. They are chemically distinct from the hosts' plant flowers scent.

Chemical profile of volatile compounds produced by rust fungus found on the barberry bush was examined for finding characteristic chemicals of rust infected leaves. The occurrence of *P. brachypodii* on barberry bushes was reviewed and discussed.



PS - poster session

**Fungi accompanying the protist *Plasmodiophora brassicae* in the soil**

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*Plasmodiophora brassicae* is a protist and a damaging pathogen of *Brassica* crops, including oilseed rape and many types of cabbages, other vegetable brassicas as well as weeds of Brassicaceae family. The pathogen is soil-borne and its resting spores are very persistent in the soil. Fields infested with resting spores can be a source of the disease even after 20 year break. The disease, called clubroot is posing a great threat to all vegetable and oilseed growing areas around the world, with European Union and Canada, as the main countries of its infection. *Plasmodiophora* is the obligate biotroph living in the cells of the host plants. Well known methods of soil fumigation and soil disinfection are unsuitable for large areas and such methods are not friendly to the environment. The aim of this study was to identify fungi associated with the presence of *P. brassicae* in selected agricultural soils with the future goal of studying their biocontrol properties. The study used Shotgun method - Illumina MiSeq approach. DNA isolation was performed using the method by Wallenhammar et al. (2012) and the method described by Zhou (1996) with modifications. Library preparation was according to TrueQuant DNA Library Preparation Kit protocol by GenXPro Trial Kit for 6 RX. Data processing was done using Kracken NGS and mapping the data against the standard database using Bowtie2 tool. The results of mapping against the standard database displayed that 97% of 10 million reads were classified as bacteria, whereas fungi accounted for 7% of the detected microorganisms. Most of them belonged to Ascomycota. The prevailing fungi were *Fusarium*, *Trichoderma*, *Purpureocillium* and *Aspergillus*. The isolation method had a significant effect on the proportions of the microorganisms studied in the sample. Many of the microorganisms preferred low soil pH, suitable for oilseed rape cultivation and the pathogen occurrence.

## MEDICAL MYCOLOGY

OS - oral session

**Alternative models to study antifungal susceptibility and virulence of pathogenic molds**

**Ulrike Binder**

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Life-threatening invasive fungal infections have increased over the last decades due to the growing numbers of patients at risk. Furthermore, fungal pathogens with low susceptibility or complete resistance to antifungal agents are emerging. In addition, the epidemiology of invasive fungal infections caused by molds has changed in the recent years and non-fumigatus, especially mucormycetes are emerging.

New tools to understand invasive fungal infections in detail and to have a screening method for novel antimycotics are required. The larvae of the greater wax moth, *Galleria mellonella*, have been widely used as alternative model system to evaluate the virulence of microbes and the efficacy of antimicrobial drugs. We have established this invertebrate model to study virulence potential of non-fumigatus molds, *A. terreus* and mucormycetes, to investigate larval immune response, test the efficacy of antifungal drugs in vivo and determine pharmacokinetic properties of the drug in the larval hemolymph.

Additionally, the successful generation of bioluminescent *Mucor circinelloides* strains, that were constructed by targeted integration of the firefly luciferase gene under control of the *M. circinelloides* promoter Pzrt1, is a first step to exploit bioluminescent imaging for in vitro and in vivo approaches. Phenotypic characteristics, virulence potential and antifungal susceptibility were indifferent to the wild-type strains and the strains were shown to be suitable to determine antifungal efficacy in vitro. The first reporter strains in a basal fungus will be further optimized and then allow real-time, non-invasive infection monitoring in insect and murine models, and the testing of antifungal efficacy by means other than survival.

OS - oral session

**Sterol composition of clinically relevant Mucorales and changes resulting from posaconazole treatment**

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In the last years the number of mucormycosis cases (also known as zygomycosis) has increased significantly. The aggressive course of the disease, delayed diagnosis and poor treatment options result in incredibly high mortality rates. Antifungals currently applied for treatment are amphotericin B, posaconazole, and isavuconazole. These drugs supposedly target either ergosterol itself or its biosynthesis, respectively, with sterol C14-demethylase (ERG11/CYP51) being the main target enzyme of the azole drugs. Nevertheless, representatives of the Mucorales may exhibit resistance to these drugs, resulting in therapeutic failure. Therefore, a full understanding of the ergosterol biosynthesis pathway and the sterol pattern of this group of pathogenic fungi is essential to determine the efficacy of antifungal therapy, to identify targets for design of novel antifungal drugs, and to investigate combinatorial effects of drugs targeting this pathway. Ergosterol biosynthesis intermediates of clinically relevant Mucorales (*Rhizopus oryzae*, *Rhizopus microsporus*) were analysed in a targeted metabolomics fashion by gas chromatography-mass spectrometry (GC-MS). The sterol content and the sterol composition of untreated hyphae were analysed and compared to hyphae confronted with sublethal concentrations of posaconazole. The azole activity resulted in significant alterations of the sterol composition, but it did not subsequently lead to a reduction of the total sterol content. The high amount of eburicol, even in untreated hyphae, let us hypothesize that these fungi could be less affected by the accumulation of non-physiological intermediates due to azole treatment, which is reflected in their lower sensitivity to azole drugs when compared to other molds.



OS - oral session

**Unravelling antibioticly active natural products of fungal endophytes from  
*Leontopodium nivale* subsp. *alpinum***

**Martina Oberhofer**<sup>1</sup>, Havva Isikoglu<sup>2</sup>, Claudia Haager<sup>1</sup>, Nadine Haubenwallner<sup>1</sup>, Judith Wackerlig<sup>2</sup>, Ernst Urban<sup>2</sup>, Martin Zehl<sup>3</sup>, Christoph Wawrosch<sup>1</sup>, Sergey B. Zotchev<sup>1</sup>

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Fungal endophytes co-exist asymptotically within living plant tissues of their host plant. Certain metabolic pathways may be outsourced to endophytes by the plant e.g. those relevant to defense, immune response or nutrient acquisition. Thus, fungal endophytes can contribute to and extend the secondary metabolome of their host.

Here, we investigated endophytes of *Leontopodium nivale* subsp. *alpinum* (Edelweiss), a traditional medicinal plant with various medicinal properties, including antibiotic activity. We harvested native alpine Edelweiss plants and isolated 859 endophytic strains, of which 55 strains were fungi. Fungal de-replication led to the identification of Ascomycota (16), Basidiomycota (2) and Zygomycota (1). Five fungi were phylogenetically distant to described species, several other fungi were never investigated for secondary metabolites. Fungal cultures were extracted with organic solvents. Total extracts were run through a pipeline of bioactivity-guided fractionation starting with antibiotic bioassays against Gram-negative and -positive bacteria, yeast and filamentous fungi. Active extracts were fractionated and fractions were tested again in bioassays. Molecular masses and sum formulae of active compounds were then determined with HR-MS analyses and compared to those of known compounds from the Dictionary of Natural Products. Putatively new compounds were subjected to NMR analysis. Antibiotically inactive fungi, mostly dark septate fungi and rare fungal taxa, were treated either with different media providing diverse sources of C and N to trigger antibiotic production, or with small molecule elicitors added to common media to activate putative silent gene clusters.

The results obtained after using these approaches will be presented and discussed.

OS - oral session

**Genetic manipulation in *Mucor circinelloides*, a model organism for mucormycosis**

**Francisco Esteban Nicolas Molina**, Maria Isabel Navarro Mendoza, Carlos Perez Arquez, Laura Murcia Flores, Carlos Lax, Pablo Martinez Garcia, Eusebio Navarro, Jose Canovas Marquez, Jose Perez Ruiz, Victoriano Garre  
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Mucorales are a group of basal fungi containing numerous species that cause mucormycosis, a fungal infection with high mortality rates. The lack of knowledge about their physiology, their high resistance to antifungals and the impossibility of using genetic transformation techniques in most of them are the reasons behind the current lack of effective treatments for mucormycosis. Fortunately, among the numerous species of the group, *Mucor circinelloides* stands as one of the few Mucorales capable of being genetically transformed, allowing the development of numerous molecular techniques for genetic and genomic functional studies in this organism. Gene silencing is one of the most studied processes in *M. circinelloides*, being responsible for a special type of antifungal resistance mediated by temporal epigenetic changes. In addition, the applied use of gene silencing has allowed the development of functional genomic techniques for the identification of new virulence factors. Along with silencing, gene disruption driven by sequence homology has also allowed us to study the particular role in *M. circinelloides* of virulence factors identified in other fungi, such as the role of a family of ferroxidases in iron uptake. The study of mutants in these genes has demonstrated their involvement in virulence of Mucorales, identifying a process of functional specialization within the components of the same gene family. Finally, the diversity of molecular techniques developed in *M. circinelloides* allowed us to study the gene response during phagocytosis. Here, we present the results of these studies, showing how *M. circinelloides* is a fundamental model for the advancement in the knowledge of Mucorales fungi, the disease they produce and the development of effective future treatments.

OS - oral session

### Use of a yeast model to improve azole drugs

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Azole drugs used to treat fungal infections are susceptible to resistance mechanisms involving (a) acquisition of amino acid changes in target sterol 14 $\alpha$ -demethylases (CYP51s), (b) active site amino acid substitutions conferring intrinsic resistance, (c) CYP51 overexpression (d) bypassing toxic metabolites using alternate pathways, and (e) drug efflux. Full-length recombinant C-terminal hexahistidine-tagged CYP51s or drug efflux pumps were functionally overexpressed constitutively from the PDR5 locus in a *S. cerevisiae* host deleted of 7 drug efflux pumps. CYP51s were also co-expressed with a cognate NADPH-cytochrome P450 reductase (NCP1) from a co-regulated locus. Phenotypic and structural analysis revealed key features of wild type and azole resistant versions of CYP51 in complex with azole inhibitors. Phenotypic studies tested for synergy with inhibitors of drug transporters.

Co-expression of CYP51s from human and agricultural pathogens with cognate NCP1s conferred enhanced CYP51 function. The tetrazole VT-1161 showed resistance conferred by overexpression of the drug target, target mutations (tyrosine [Y] to phenylalanine [F] in the active site) and overexpression of drug efflux pumps similar to fluconazole and voriconazole. The Y to F mutation, which alters a water-mediated hydrogen bond network in the active site, was overcome by modifying the linker between the head and tail of the VT-1161. Inhibitors of drug efflux showed VT-1161 is susceptible to drug efflux by *C. albicans* CDR1.

The recombinant CYP51s facilitate structure-directed discovery of inhibitors not susceptible to particular CYP51 resistance mutations. Recombinant drug efflux pumps show how inhibitors of drug efflux can block azole resistance.

OS - oral session

### **Bioactive compounds from Northeastern Thailand**

**Pathompong Paomephan**<sup>1</sup>, Benjarong Thongbai<sup>2</sup>, Kevin Becker<sup>2</sup>, Chuenchit Boonchrid<sup>1</sup>, Marc Stadler<sup>2</sup>

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The discovery of new antibiotics is badly needed to combat the constantly increasing number of multi-drug-resistant microbes. Several known antibiotics are derived from fungi, and the last class of antibacterial drugs that reached the market was actually derived from cultures of a mushrooms belonging to the Basidiomycota (1). Since many Basidiomycota produce antibiotics as a means of chemical defense or to replace other species by antagonism in their natural habitat, several unique secondary metabolites with prominent biological activities have already been reported from these organisms (2). In this work, 109 strains of Basidiomycota were collected from Northeastern Thailand and cultured. They were obtained to screen at primary screening using agar diffusion technique and dual antagonistic test against bacteria, yeast and filamentous fungi. 18 strains were shown to have an antibacterial activity. Activity against filamentous fungi was noted in 29 strains and 4 strains exhibited anti-yeast activity. These strains were subjected to small scale cultivation in different culture media and the resulting organic extracts checked by HPLC-DAD/MS to evaluate their secondary metabolite profiles. Then, scale-up-cultivation was done to produce larger amounts of crude extract for fractionation using preparative HPLC. Antimicrobial activity of each fraction was checked using a serial dilution assay to obtain the minimum inhibitory concentration (MIC). The active principles were isolated to purity and their structures are presently being elucidated using NMR spectroscopy and high resolution mass spectrometry. The most important results of the project will be reported.

PS - poster session

**Intrinsic short-tailed azole resistance in mucormycetes**

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**Aim.** Mucormycetes are intrinsically short-tailed azoles resistant. We used the high resolution X-ray crystal structure of *S. cerevisiae* lanosterol 14 $\alpha$ -demethylase (LDM) in complex with voriconazole (VCZ) to postulate that the amino acid substitution Y129F in *Rhizopus arrhizus* LDM (RaLDM) F5 is associated with their intrinsic resistance. Our aim was to experimentally test this hypothesis.

**Methods.** Full-length recombinant C-terminal hexahistidine-tagged LDM (F1 and F5) of *R. arrhizus* were overexpressed from the *PDR5* locus in a *S. cerevisiae* host lacking 7 drug efflux pumps. Their cognate NADPH-cytochromeP450 reductase (RaNCP) was overexpressed from the *PDR15*. Phenotypic and biochemical analysis were used to determine structural and functional features of RaLDMF1, RaLDMF5, without and with co-expression of RaNCP.

**Results.** RaLDMF1 and F5 were functionally overexpressed in *S. cerevisiae*. RaLDMF5 appeared to be the major contributor to azole resistance, with its expression resulting in a 32x higher VCZ minimal inhibitory concentration (MIC) than the recipient strain. Co-expression of RaLDMF5 with NCP resulted in additional 4-fold increase of VCZ MIC when compared with RaLDMF5 single expression. MICs for long-tailed azoles were unaffected by the LDM supplementation. *S. cerevisiae* co-expressing RaLDMF1 with NCP had similar MIC values as the recipient strain. Azole-susceptibility patterns of *S. cerevisiae* co-expressing RaLDMF5 and RaNCP closely matched those of *R. arrhizus*. Phenotypic analysis of recombinant LDMs expressed in *S. cerevisiae* provides insights on the relevance of mutations located in the substrate binding pocket. This system allows assessment of effects on the drug target without interference from drug efflux pathways.

**Conclusion.** LDMF5 was found to be the dominant isoform in *R. arrhizus* that confers short-tailed azole resistance. Biochemical and structural analysis of the purified protein are expected to provide deeper understanding of the observed phenotypes.

PS - poster session

**Mycelial cultures of *Lentinula edodes* enriched with bioelements as a potential anti-inflammatory material**

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*Lentinula edodes* is one of the most popular edible mushroom species. It is obtained by cultivation and under biotechnological controlled conditions. Its fruiting bodies are of both dietary and therapeutic importance. The most popular metabolites are  $\beta$ -glucans (e.g. lentinan registered as a drug), ergothioneine, lovastatin, phenolic derivatives, bioelements, and vitamins. In addition, the culture mycelium of *L. edodes* is capable of accumulating Cu, Zn and Se, which play a key role as anti-inflammatory factors.

Due to these advantages, *L. edodes* was chosen for the research. The aim of the study was to obtain mushroom biomass rich in bioelements, which may present anti-inflammatory properties. The extracts were prepared from biomass derived from in vitro cultures of *L. edodes*. Cu and Zn ions in the form of inorganic and organic compounds, and selenium in the form of Selol 5% (preparation containing 50 mg/mL of Se(IV) triglycerides), were added to the modified Oddoux medium in which *L. edodes* was cultured. Mycelium grown on Oddoux liquid medium served as controls. The anti-inflammatory properties of *L. edodes* mycelium were evaluated in lipopolysaccharide-activated RAW 264.7 cells using Western blot technique.

On the basis of the levels of COX-2, Nrf2, PPAR $\gamma$ , and FABP4 determined using Western blot technique, it was found that the addition of Cu, Zn or Se compounds enhanced the anti-inflammatory properties of *L. edodes* mycelial extracts. The addition of copper in both form (copper sulfate, copper gluconate) proved to be the most beneficial. This indicates that *L. edodes* mycelium cultured on a suitable medium may be used as a potential component of natural anti-inflammatory preparations.

Source of funding: National Science Centre, Poland 2017/25/N/NZ7/00554

PS - poster session

**Role of dephosphorylation of dolichyl diphosphate in protein glycosylation and morphological transitions in *Candida albicans***

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The essential role of dolichyl phosphate (DolP) as a carbohydrate carrier during protein N-glycosylation is well established. The cellular pool of DolP is derived from the de novo synthesis in the dolichol branch of the mevalonate pathway and from the recycling of DolPP after each cycle of N-glycosylation, when the oligosaccharide is transferred from the lipid carrier to the protein and DolPP is released and then dephosphorylated. In *Saccharomyces cerevisiae* the dephosphorylation of DolPP is known to be catalyzed by the Cwh8 protein. To establish the role of the Cwh8p orthologue in another distantly related yeast species, *Candida albicans*, we studied its mutant devoid of the CaCWH8 gene. As in *S cerevisiae*, the mutant was impaired in DolPP recycling. This defect, however, was accompanied by an elevation of cis-prenyltransferase activity and higher de novo production of dolichols. Despite such compensatory effect protein glycosylation, cell wall integrity, filamentous growth and biofilm formation of the mutant were impaired. These results suggested that the observed effect is not due to the lack of DolP for the N-glycosylation process. Activity of oligosaccharyltransferase could be inhibited by DolPP which is not utilized in this mutant.

PS - poster session

### **Dermatophytes isolated from wild rodents**

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Spectrum of skin pathogens from the group of dermatophytes (Onygenales) is well studied in livestock/pet animals and men due to their economical and public health importance. In contrast, spectrum of dermatophytes in wild animals is almost unknown, despite the possible transmission to humans. Especially rodents are suspected to be important reservoir of many dermatophytes (e.g. *Trichophyton mentagrophytes*, *Arthroderma curreyi*, *A. quadrifidum*) due to their occasional isolation from these animals. The aim of this study was to comprehensively investigate the spectrum of dermatophytes in rodents. Samples from wild African and Czech rodents were collected by brushing their coat and cultivated using selective medium (SGA with cycloheximide and chloramphenicol) and used for DNA extraction and NGS libraries preparation. We determined several species of dermatophytes which all are considered as geophilic (soil reservoir). However, frequent isolation from rodents indicate that this categorisation may be not strict. Some of these species are found in clinical material causing severe infections in weakened host. On the other hand, zoophilic dermatophytes (e.g. *Trichophyton mentagrophytes*, *Microsporum canis*) previously reported from rodents were absent in our data. Isolates acquired in this study will be compared with closely related strains previously obtained from other animals and clinical samples. This might help us to prove where is the reservoir located and to understand the way of transmission.



PS - poster session

***Cryptococcus* species complex stands out among yeast-like fungi by the ability to form Titan cells in vitro**

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*Cryptococcus* species complex is unique for the number of virulence factors that allowed these species to evolve into human pathogens. One example is a unique morphological transition to form enlarged Titan cells that escape phagocytosis by host immune cells. Titans, 2-20 times larger than normal cells, generate haploid or aneuploid daughter cells that exhibit resistance to physico-chemical factors and antifungal drugs, which further contributes to infection. It remains unclear whether non-*C. neoformans*/non-*C. gattii* basidiomycetous species are capable of titanization. Based on a recently published in vitro protocol, we performed a survey of several basidiomycetous and ascomycetous yeasts and propose that titanization is unique to *C. neoformans* and *C. gattii*. In addition, we find that under in vitro titanization conditions, fetal bovine serum (FBS) possesses activity that inhibits growth of *C. gattii* and kills *C. neoformans* if incubation is conducted in the absence of 5% CO<sub>2</sub>. Furthermore, lowering of pH or addition of an antioxidant, rescues growth in the presence of FBS and allows titanization even in the absence of 5% CO<sub>2</sub>. We also find that the inhibitory/killing activity of FBS requires low cell density. Moreover, our results involving acapsular mutants suggest that capsule plays a role in FBS-mediated killing. Currently studies are ongoing to test the role of quorum sensing and lysozyme in FBS-mediated growth inhibition. We are also currently testing *Cryptococcus amylolentus* and chosen species of *Kwoniella* genus, which are closely related to *Cryptococcus* species complex. Our data implicate titanization as a unique feature of *C. neoformans* and *C. gattii* and provide novel insights into the nature of this unusual morphological transition.



PS - poster session

**Antitumor effect of the nigerooligosaccharides obtained by acid-hydrolysis of  $\alpha$ -(1→3)-glucans from fruiting bodies of *Fomitopsis betulina***

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The fruiting bodies of *Fomitopsis betulina* can be used in some fields of modern medicine. The fungus has a long tradition of being applied in folk medicine as an antimicrobial, anticancer, and anti-inflammatory agent. Biologically active compounds such as triterpenoids have been isolated. The mushroom is also a reservoir of valuable enzymes and other substances such as cell wall polysaccharides.

Nigerooligosaccharides, a mixture of (1→3)- $\alpha$ -linked D-glucose oligosaccharides, were prepared by partial acid hydrolysis of  $\alpha$ -(1→3)-glucans isolated from *Fomitopsis betulina* fruiting bodies and analyzed by HPLC and MALDI-TOF Mass Spectrometry. Further, their anti-cancer potential was evaluated in in vitro assays in a colon cancer cell model.

Oligosaccharides are carbohydrates widespread in nature. Besides the natural source, the production of new oligosaccharides is currently attractive to pharmaceutical and food industries.

In the present study, anticancer activity of nigerooligosaccharides obtained from *Fomitopsis betulina*  $\alpha$ -(1→3)- glucans, were tested in in vitro model. The tested oligosaccharides showed antiproliferative (MTT assay) and pro-apoptotic (Annexin V-FITC and PI technique) features against colon cancer but not against normal epithelial colon cells. Additionally, we did not observe cytotoxic activity (neutral red and lactate dehydrogenase assays) of nigerooligosaccharides against several types of normal cell lines. Our study supports the opinion that hydrolyzate of  $\alpha$ -(1→3)- glucans may be used as diet or therapy supplements.

PS - poster session

**Potential of *Aspergillus* proteases usage in biomedicine**

**Svetlana Timorshina**, Anna Rakova, Aleksandra Ochneva, Daria Bednenko, Alexander Osmolovskiy  
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At the present time, biomedicine is an advanced branch of science, allowing to develop new methods of treatment and diagnosis of many diseases, including diseases associated with dysfunction of the hemostasis system, and biofilm-associated infections. Micromycetes have great potential as producers of enzyme components of pharmacological preparations. It is known that filamentous fungi of the genus *Aspergillus* synthesize a wide range of extracellular proteases with different substrate specificity, and therefore screening was conducted among members of this genus to identify proteases with activities significant for medicine. It has been shown that micromycetes *A. oryzae* and *A. sclerotiorum* produce exoproteases capable of activating protein C, a key factor in the human anticoagulant system. These proteolytic enzymes with high activity against protein C (31.4 and 35.5  $\mu\text{moles pNA} / (\text{ml} \times \text{min})$  respectively) can replace an expensive analog derived from snake venom and used as part of diagnostics to determine the amount of protein C in human blood plasma. It was also shown that *A. flavus* synthesize a highly active protease capable of cleaving fibrin (39.8  $\mu\text{moles pNA} / (\text{ml} \times \text{min})$ ), which can be used in the treatment of thrombosis. Recently, the search for enzymes that lyse the biofilms of microorganisms is of high interest, since they are not only a factor in the deterioration of industrial equipment, but also the cause of the development of many diseases. So, *A. alliaceus* proteases are able to reduce the number of metabolically active cells in the biofilm of *E. coli* K12 by 20%. Thus, the study of the proteases of filamentous fungi is very promising from the point of view of biomedicine.

PS - poster session

**Effectiveness of mycostats and disinfectants on yeast strains isolated from tap water**

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Numerous studies have shown that fungi are commonly found in water distribution systems, in drinking and pool water. The reduced sanitary quality of water is directly related to the problem of contact between contaminated water and mucous membranes of people using such facilities. Treatment of skin symptoms that appear after infection is problematic due to the lack of a universal antifungal agent. The aim of the study was to evaluate and compare the effectiveness of mycostats and disinfectants on yeast strains isolated from tap water during previous own research.

28 yeast strains isolated from tap water and 4 reference strains from the national collection were analyzed. Fungi are classified into 10 genera, including *Candida*, *Cryptococcus*, *Exophiala*, *Saccharomyces* and *Saccharomycopsis* belonging to potential pathogens. To test the effectiveness of products generally available on the market, with mycostatic and/or mycocidal effect, preparations were selected in various forms: cream, gel, liquid and oil. These were respectively: clotrimazole, miconazole, resorcinol and tea tree oil. The susceptibility tests were carried out using the well-diffusion method on Sabouraud agar without antibiotics.

Significant differences in yeast susceptibility to the agents used were indicated. The strongest effect was caused by miconazole and clotrimazole. These preparations were active against 100% strains. A larger percentage of species (82%) showed reduced sensitivity or even high resistance to the tested medicament (e.g. *Candida albicans* - the most common etiological agent of fungal infections in humans). The weakest effects were tea tree oil and resorcinol. This clearly indicates a weaker effect of antiseptics on yeast growth than antifungal medicines.

PS - poster session

### **Mold fungi isolated from the sports locker room**

**Tomasz Balabański**, Anna Biedunkiewicz

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Increased physical effort and associated periodic resistance fluctuations may increase the body's susceptibility to fungal infections. Among the factors that increase the risk of mycosis of the skin surface, among other, amateur or competitive sports are mentioned. Therefore, athletes are particularly exposed to direct contact with indoor mold fungi.

The aim of the study was to evaluate the taxonomic diversity of mold fungi occurring in sports locker rooms and toilets, located in school buildings and sports halls, including the Biosafety Level (BSL) of isolated fungi.

The experimental material was collected by surface swabbing from walls, shower trays, showers and other sports locker equipment at volleyball halls in the city of Olsztyn (NE Poland).

From 48 collected samples, a total of 52 strains of mold fungi were obtained, in which 18 species were found, classified into 10 genera. The most numerous were: *Aspergillus fumigatus* (21.15%), *Phoma eupyrena* (17.31%), *Aspergillus niger* and *Phoma minutispora* (11.54% each). Three species: *Aspergillus fumigatus*, *Acremonium kiliense* and *Aspergillus flavus* were classified to BSL-2, which constituted 16.67% of all isolated species. Most fungi were isolated from the locker rooms walls (40.38%), slightly less from showers (36.54%). The shower trays surfaces were the least contaminated (23.08%).

The conducted research clearly indicates a high degree of contamination of sports locker rooms and toilets with mold fungi and suggest the necessity of conducting regular mycological monitoring of this type of rooms, due to the health of users.

PS - poster session

**Two poorly defined populations revealed by microsatellite analysis of *Trichophyton rubrum* complex**

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*Trichophyton rubrum* is one of the most common pathogens worldwide causing predominantly skin and nail but infections in humans. Due to its high importance, this species was extensively studied, and many taxa were proposed based on phenotypic differences. Most of taxa were with *T. rubrum* synonymised based on the phylogenetic analysis results but many others are still recognized by some authors (e.g. *T. violaceum*, *T. megninii*, *T. soudanense*). A total of 121 morphologically atypical *T. rubrum* complex isolates corresponding to different varieties/species were selected. These strains were obtained from Czech patients between 2011-2015 and analysed using 10 microsatellites markers (Graser et al. 2007, Gong et al. 2016) together with the ex-type isolates of 11 species belonging to *T. rubrum* complex. Two major populations corresponding to *T. rubrum* and *T. violaceum* species were recognized based on the STRUCTURE analysis. However, many genetically transitional strains were detected forming a genetic continuum between these populations/species. These intermediate strains do not originate from introgressions but they seem to be result of randomly generated clones. Genetic, morphological and clinical differences among *T. rubrum* complex strains can be most probably explained by the existence of local populations of fungus or specific behaviour of patients. For example, explanation why strains of *T. rubrum* complex strains isolated in Europe cause rather nail and skin infections but tinea capitis in Africa, do not necessarily indicate that they should be considered separate species.

PS - poster session

**Differential gene expression of species from *Trichophyton benhamiae* clade**

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Species from *Trichophyton benhamiae* clade (Onygenales) are closely related skin pathogens of human or pet animals with frequent transmission to humans. Nowadays, the clade become more important due to emerging epidemic of species *T. benhamiae* amongst children in Central Europe. This species has the same main host (guinea-pigs) and geographical distribution as other two species from the *T. benhamiae* clade. However, *T. benhamiae* is more infectious or more successfully transmitted from guinea pigs to humans. This phenomenon can be explained by different virulence factors. Unfortunately, not much is known about the overall picture of the infection process, although few virulence factors have been identified for *T. benhamiae* clade, such as subtilisins, fungalysins, hydrophobins and others. Therefore, we compared their gene expression during infections to find a potential virulence factors and finally design RT-PCR probes for those species. Differential gene expression of 5–8 strains from each species from *T. benhamiae* clade has been studied. Strains were cultivated on artificial media (with or without keratin) and compared with cultures cultivated on murine skin grafts. For the purpose of this study, we successfully modified the murine skin grafts cultivation protocol and process of obtaining RNA from skin samples. Differences between transcriptomes of dermatophytes might provide us insight into this complex problematic. Moreover, the outcome of this study, namely the discovery of new virulence factors might be useful for development of antimycotic treatment.

PS - poster session

**Characterization of bioactive compounds and antioxidant activity of some medicinal macromycetes harvested from different regions of Armenia**

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Many fungal strain's extracts show strong antioxidant activity what are comparable to classic antioxidants such as Vitamin C or E. In our current research we have investigated some major compounds responsible for antioxidant activity in 10 different species of fungi (*Hypholoma fasciculare*, *Agaricus bisporus*, *Pleurotus ostreatus*, *Trichaptum abietinum*, *Polyporus squamosus*, *Schizophyllum commune*, *Calvatia gigantea*, *Armillaria mellea*, *Scleroderma verrucosum*, *Trametes versicolor*). All fungi were harvested in June-August 2018 from different regions of Armenia. Antioxidant activity was assessed via potentiometric method based on oxidation/reduction of Fe<sup>2+</sup>/Fe<sup>3+</sup>. Phenolic, flavonoid and carotenoid contents of fungi were determined by spectrophotometry. Thin-layer chromatography was performed as well to characterize flavonoids. Results indicate that all investigated species show antioxidant activity except *Trichaptum abietinum*. *Agaricus bisporus* and *Pleurotus ostreatus* show highest antioxidant activity among these species. Phenolic compounds have a significant role but there are other antioxidants present as well. Despite spectrophotometric analysis of flavonoids showed high content of them in many fungal extracts, thin-layer chromatography showed no flavonoid presence in those extracts. We believe that due to many sterol compounds present in fungi what are absent in plants the spectrophotometric analysis showed false positive results so it is not recommended to use for fungal extracts. *Scleroderma verrucosum* has the highest phenolic content. *Hypholoma fasciculare* has highest carotenoid content, *Schizophyllum commune* and *Polyporus squamosus* contain trace amounts of carotenoids. Further researches of antioxidant activity on lipid membranes and biological objects are in progress.





PS - poster session

**Elucidating species boundaries between agents of superficial mycoses *Trichophyton interdigitale* and *T. mentagrophytes***

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Dermatophytes are fungi which cause mycotic infections of skin and its derivatives. *Trichophyton interdigitale* is one of the most common agents of mycotic infections in humans. While closely related species *Trichophyton mentagrophytes* is predominantly the agent of superficial skin infections in domesticated animals but can be also easily transmitted to human and cause skin lesions. Due to different source of infection, the differentiation of these species is clinically and epidemiologically significant.

These species should be distinguishable based on characteristic phenotypic features, including macromorphology of colonies, presence of macroconidia and spiral hyphae, etc. However, recent studies indicated that the correlation between clinical picture of infection, and phenotype and genotype of the pathogen are not as clear as expected. The species boundaries between these species need to be re-evaluated using molecular and phenotypic methods. The aim of this study is to find species limits between *T. interdigitale* and *T. mentagrophytes*. The alternative hypothesis is that these two species represent in fact only one species with a broad host range and variable phenotype. 120 isolates identified as *T. interdigitale* / *T. mentagrophytes* were obtained from patients. The analysis of micro- and macromorphology, and physiology was performed together with molecular characterization of the strains by DNA sequences from three loci: ITS rDNA,  $\beta$ -tubulin and translation elongation factor 1- $\alpha$  (TEF). Two species delimitation methods were compared: genealogical concordance phylogenetic species recognition (GCPSR) and methods based on the multispecies coalescent model. Correlation between phylogeny, clinical manifestation and phenotypic data was performed.



PS - poster session

**Contents of heavy metals, macro- and microelements in selected species of edible mushrooms in NW Poland**

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The aim of the study was to assess the accumulation capacity of selenium and other selected microelements, macroelements and heavy metals by *Boletus edulis*, *Imleria badia* and *Leccinum* spp. on the background of habitat conditions in Western Pomerania (NW Poland). Concentration of selenium in dried mushrooms was determined using the Watkinson's spectrofluorometric method, modified by Grzebuła and Witkowski. Atomic absorption spectrometry was performed with a Unicam Solar 929 device to identify the micro- and macroelements dissolved in 0.5 mol dm<sup>-3</sup> HCl, as well as the total content of micro- and macroelements dissolved in the mixture of concentrated acids HNO<sub>3</sub>+HClO<sub>4</sub>; phosphorus was determined colorimetrically. The examined species grew on the organic layer of sandy soils. The soils were strongly acidic, rich in exchangeable P, poor and medium rich in exchangeable K and poor in exchangeable Mg. The soils were not contaminated with heavy metals (Cu, Cd, Pb and Zn), only in some cases an increased level of Ni was found. The research showed that *Boletus edulis* contains significantly more Se and Mg than *Imleria badia* and *Leccinum* spp. All the tested species did not show significant differences in Ca, K, Mg, P, Na, Cu, Zn, Ni, Cd, Pb, Mn and Fe content. The content of Se in our samples was significantly positively correlated with the content of Zn, Mn and P in soil, and negatively with the content of K and Mg. In the result, we found that the more acidic and richer in organic matter the soil is, the higher is the soil proportion of selenium, which is however not reflected in the content of Se in fungal tissues.

PS - poster session

**Yeast *Saccharomyces cerevisiae* as an eukaryotic model to study genotoxicity of potential drugs**

**Magdalena Cal**<sup>1</sup>, Irwin Matyjaszczyk<sup>1</sup>, Ireneusz Litwin<sup>2</sup>, Daria Augustyniak<sup>1</sup>, Grażyna Majkowska-Skrobek<sup>1</sup>, Rafał Ogórek<sup>1</sup>, Young Ko<sup>3</sup>, Peter Pedersen<sup>4</sup>, Stanisław Ułaszewski<sup>1</sup>

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*Saccharomyces cerevisiae* is commonly used as an eukaryotic model to study of cell cycle regulation, cellular response to DNA damage and genome stability mechanisms. *S. cerevisiae* is also a useful model to investigate biological activity of novel chemotherapeutics and the cellular response to their presence. 3-bromopyruvate (3-BP) is an analog of pyruvic acid that exhibits strong anticancer activity. In the cell 3-BP becomes highly reactive, showing strong alkylating properties towards proteins. It was shown that 3-BP may inhibit key glycolytic enzymes and affects mitochondrial functions. However, the exact mechanism of 3-BP action is still under debate. Here we investigated if 3-BP is a genotoxic agent. Our results indicate that in yeast 3-BP induces DNA damage mostly in the form of DSBs probably as a result of generation of high levels of ROS. We found that like MMS, 3-BP triggers H2A-P during DNA replication but is unable to induce DNA damage checkpoint outside of S-phase. We also found that neither BER (base excision repair) nor NER (nucleotide excision repair) or NHEJ (non-homologous end joining) mutants were more sensitive to 3-BP than wild type (WT). On the other hand strains lacking RAD52 and RAD59 genes, that are crucial for homologous recombination, showed increased sensitivity to 3-BP. Taken together, our data imply that 3-BP displays genotoxic activity suggesting that 3-BP-dependent DNA damage may be, at least in part, important for anticancer activity of this drug. Research focused on 3-bromopyruvate are extremely important, because in the future they may lead to more effective anticancer and antifungal therapies.

PS - poster session

**Chloroquine & Primaquine Diphosphate as Possible Alternative Drugs for the Treatment of Cryptococcal Infection**

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This study investigates the action of chloroquine (CQ) and primaquine (PQ) diphosphate on the growth of *Cryptococcus*, which is a pathogen responsible for cryptococcal meningitis, one of the leading causes of mortality in persons living with HIV/AIDS. The direct effect of CQ and PQ on cryptococcal isolates showed a distinctive growth reduction ranging from 50 - 89% ( $p < 0.01$ ) and 38 - 74% ( $p < 0.01$ ) respectively, with CQ exerting a greater inhibitory effect. The minimum inhibitory concentrations (MICs) were determined to be 50 $\mu$ M for CQ and 60 $\mu$ M for PQ. Their MICs inhibited *Cryptococcus* by adversely affecting the mitochondrial membrane potential (MMP) with CQ and PQ accounting for 43 - 60% ( $p < 0.01$ ) and 52 - 65% ( $p < 0.01$ ) respectively. Hence, the loss of MMP resulted from a significant dislodging of cytochrome c ( $p < 0.05$ ) from the mitochondria and overproduction of reactive oxygen species ( $p < 0.01$ ). CQ and PQ also induced disruption of the cell wall integrity by creating pores ( $p < 0.05$ ), causing leakage of cellular components ( $p < 0.05$ ) and led to cell death. Both drugs did not negatively affect macrophages (host) as we recorded a broad therapeutic index of 20:1. Remarkably, CQ and PQ significantly enhanced the phagocytic efficiency of macrophages against *Cryptococcus* by a maximum of 27% ( $p < 0.05$ ) and 32% ( $p < 0.05$ ) respectively at  $\frac{1}{2}$  their MICs. Based on this study, CQ and PQ can serve as candidate drugs to control the growth of *Cryptococcus*.

## FUNGAL DIVERSITY



OS - oral session

**Keeping an eye on what we, literally, mostly cannot see: fungal diversity and the identification of “bellwethers” of global change**

Carrie Andrew

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Ironically, the complexity in the ways which fungi live can impede our ecological understanding of their biodiversity, because their roles and interactions in the natural environment are so varied and, mostly, cryptic. Information on fungal diversity is critical to planning and mitigating potentially negative consequences of global change, and especially related to conservation. Climate and land-use change are, broadly, highly influential. In this talk, analyses are discussed from research conducted via museum and citizen science fruit body recordings, integrated with open-source environmental and reproductive traits data. There is substantial likelihood that further climatic change will influence fungal diversity, as evidenced by contemporary impacts to it, as well as, the phenology, assembly, and distributions of fungi in Europe. These consequences of global change are discussed with a mind towards moving fungal conservation forward, in this respect by identifying taxa that may be, in terms of their fruiting, either especially sensitive or resistant to climate change. The ecophysiology of fungal reproduction, at the macroecological scale - i.e., identifying “the bellwethers” of global change - can perhaps be used as an assessment technique for conservation monitoring of a group of organisms that are so vastly diverse, that to observe them all remains, literally, a challenge that we must continue to understand.

OS - oral session

**DNA-barcoding of fungi in Austria - state of the art of the HSRM project at the University of Vienna**

**Irmgard Krisai-Greilhuber**

*University of Vienna, Vienna, Austria*

A considerable number of sequences of Austrian fungi can be found in publicly accessible databases (GenBank, UNITE), but without being marked as or meeting the quality requirements of DNA-barcodes. To improve this situation, a three-year higher education structure funding project (HSRM) is currently underway within the Austrian DNA barcoding initiative ABOL with the aim of establishing barcoding pipelines at Austrian universities. ABOL is a national network of institutions and experts dealing with research on Austrian biodiversity. It aims to generate barcodes of all species of animals, plants and fungi occurring in Austria. The DNA barcodes will be publicly available. Designed as a long-term initiative the overall ABOL project started in 2017 succeeding a pilot phase. The coordination is done by the Natural History Museum Vienna and financed by the Austrian Federal Ministry of Education, Science and Research. Barcodes will be generated in subprojects assigned to group-specific clusters. In some groups species counts are rough estimates as no recent data are available. ABOL will contribute substantially to a reliable data basis. From 2017-2019 the University of Vienna will contribute ITS sequences of macromycetes and animals. In the first two years about 3000 barcodes of about 1700 fungal species could be achieved. The fungarium WU-Mykologicum at the university is the central resource for the samples. Some results in the genera *Cortinarius* and *Entoloma* a.o. are presented, such as new species, new records for the area or rediscovery of red list species, clarification of incorrect determinations and uncertain interpretations. Barcoding of fungi is now successfully established at the University of Vienna and will bring further interesting results in the next few years.



OS - oral session

**Folk knowledge and observations as new sources of information about the ecology of macrofungi and changes in local mycobiota**

Marcin Kotowski, Łukasz Łuczaj

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This field study is the first detailed documentation on the use and knowledge of fungi in any large mycophilous region of Europe. A total of 695 semi-structured interviews were carried out among local informants in 38 localities proportionally distributed throughout the study area (one locality approximately every 30 km). Interview questions included which fungi species were collected, what their perceived habitats were, and whether any changes had been noted in their abundance. Altogether, 92 taxa identified to the species or genus level were recorded, among which 76 species were used as food, 21 taxa were known as toxic, and 11 taxa used for non-culinary purposes. Out of 76 identified edible fungi species, 47% (36 species) were identified using the ITS DNA barcode method. Eleven of them were identified exclusively by molecular analysis. Two species new to the mycobiota of Poland, *Hydnum ellipsosporum* and *Paxillus cuprinus*, were found. Out of 695 respondents, 366 (53%) noticed a steady decrease in macrofungi abundance during their lifelong observations. 13% of respondents observed abundance fluctuations, but assigned this phenomenon to natural changes related to differences in seasonal rainfall and temperature distribution. 34% of respondents did not notice any abundance changes and only 0.14% (one person) claimed to have observed a steady increase in local macrofungi abundance. The only species with increased abundance, noticed by fifteen independent respondents, was *Imleria badia*. 186 respondents (27%) claimed that the possible reason behind the steady decrease in macrofungi population was "drought". This collected information on the ecology of fungi proves that traditional knowledge does not generally diverge from scientific knowledge related to this topic.



OS - oral session

**Soil fungal communities in industrial pollution gradients: good models for testing community assembly hypotheses**

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In recent decades, phylogenetic diversity (PD) became one of the primary descriptors of biotic communities in conservation biology, evolutionary biology, and ecology. However, its interpretation and applicability for the analysis of community assembly is hotly debated. In the era of high-throughput sequencing, multispecies soil fungal communities are very informative from the PD perspective compared to many macroorganisms which are usually represented by few species per sampling unit. Therefore, well designed and extensive analyses of soil fungal PD can be powerful in testing the main theories of community assembly.

In present work, by means of next-generation sequencing we inventoried fungi in 180 soil samples collected along two 30 km pollution gradients caused by large copper smelters operating for 74 and 104 years. The gradients stretch through spruce-fir forests in the Middle Urals and birch-pine forests in the Southern Urals. Such large-scale pollution gradients are good experimental systems for the exploration of how environmental filtering transforms the phylogenetic structure of biotic communities.

The sequencing has yielded more than 9.5 million of high-quality fungal reads which were grouped into 5491 operational taxonomic units and mapped on the phylogenetic tree using hybrid-tree approach (Ghost-tree). Based on this data we estimated various PD metrics. Depending on taxonomic level and fungal group we revealed different effect of pollution on fungal PD (from phylogenetic overdispersion to clustering). We explain the results in the context of fungal trophic mode, sensitivity to pollution, and methodology of PD analysis.

The study was supported by Russian Foundation for Basic Research (projects 19-04-00921 and 18-29-05042).

OS - oral session

### **How to estimate fungal diversity in bogs?**

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Fungi play a significant role in carbon cycling in peatland ecosystems as mycorrhizal symbionts, saprophytes, endophytes or parasites. Although there are many studies and local surveys focused on macromycetes of peatlands, the diversity and structure of whole fungal communities is still insufficiently known. Current e-DNA methods are promising tool for a diversity assessment and monitoring of fungal community response to disturbances or restoration efforts. We used our long-term monitoring plots (3 x 3 m) in the Jizerské Hory Mts (Czech Republic, Central Europe), to find out (1) how data on fruit bodies agree with e-DNA data and (2) which part of diversity is hidden when conventional soil core sampling (peat and mixed litter) is applied instead of sequencing of individual substrates. In total, we detected 68–103 OTUs per plot, of which only 14–36% were revealed in soil core samples. A poor correspondence between the recorded OTUs and both visually inspected fruit bodies (31 species) and the literature reports on fungal taxa associated with the analysed substrates was observed. Fungal assemblage was mostly influenced by substrate identity, whereas the effect of site was minimal. Only a few OTUs behaved as generalists. The most specific fungi were found in dead parts of *Calluna vulgaris*, *Trichophorum caespitosum* and *Drosera rotundifolia*. Such high substrate heterogeneity prevents the sequencing of soil core samples for an exhaustive inventory of fungal diversity in bogs.



OS - oral session

**Effects of macroclimate and resource on the diversity of tropical wood-inhabiting fungi**

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Wood-inhabiting fungi are one of the most important groups of organisms as they contribute substantially to carbon and nutrient cycles by decomposing dead wood. Current knowledge of their occurrence, distribution, and drivers of their diversity derives almost exclusively from temperate and boreal forest ecosystems. We sampled wood-inhabiting fungi across Benin, a tropical country in West Africa with a strong north–south seasonality gradient consisting of three macroclimatic zones. We aimed at determining whether macroclimate or the resource (size or amount of dead wood, number of host tree species, and stage of wood decomposition) is more important for their diversity. Variation partitioning revealed a stronger partial effect of resource on fungal species richness and a strong effect of macroclimate on the community composition. A more detailed linear mixed-effects model revealed a significantly positive effect of host richness, amount of dead wood, and macroclimate on fungal species richness and a significantly positive effect of macroclimate and stage of wood decomposition on the community composition. These findings are consistent with patterns found in temperate and boreal ecosystems, which indicates the existence of general pattern of the diversity of wood-inhabiting fungi. Based on these results, we recommend that existing knowledge should be exploited for the conservation of wood-inhabiting fungi in tropical Africa.



OS - oral session

**Current climate change and distribution patterns of clavarioid fungi (Agaricomycetes) diversity in the Eurasian Arctic**

**Anton Shiryaev**

*Institute of Plant and Animal Ecology UrB RAS, Ekaterinburg, Russia*

A comprehensive study of the dynamics of climate change for flora and fungal communities in high latitudes of the planet is currently relevant because of climate changes and anthropogenic impact. Contemporary climatic factors (mean annual temperature and precipitation, permafrost level, etc.) are statistically important for determining the current level of species richness of fungi. On the other hand, there is no relationship between the species richness of fungi in localities and the richness of the flora, as well as the level of soil pH. It has been established that with the rising temperature in the Arctic, permafrost recedes, flora gets more «boreal», timberline moves to the north, and new typical for the forest zone species of fungi appear in the Arctic. We collected large-fruited fungi on substrates previously absent in the region 20-50 years ago: a large fallen woods, boreal mesophilic herbs, rich soil and litter. Many of such fungi occur now further to the north, and their distribution is limited only by the lack of the required substrate (and not by the climatic factor). On the other hand, fungi with Arctic and Alpine distribution retreat to the north, their numbers gets declined. Introduced plants to the gardens and parks of northern cities, as well as firewood and building materials compound potential suitable grounds for alien fungi in the Arctic regions. As a result of a 20-year study, based on an analysis of the species richness in 86 localities (each 100 km<sup>2</sup>), a map of the species richness distribution of clavarioid fungi of high latitudes of Eurasia was compiled.

Work was supported by RFBR (project № 18-05-00398).



PS - poster session

**Distribution of microfungi through soil profiles at the Hallamish dunefield, Negev Desert, Israel**

**Isabella Grishkan**<sup>1</sup>, Giora Kidron<sup>2</sup>

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We examined the depth-wise distribution of microfungi through 0-50-cm of crusted sandy and playa profiles, and through 0-40-cm of sandy profiles under bare surface and different shrub species at the Hallamish dunefield in the Negev Desert, Israel. A total of 224 species from 82 genera was isolated using the soil dilution plate method. High load of solar radiation, high temperatures, and low moisture (at the surface) as well as strongly limited aeration, water infiltration, and increased salinity (at the playa depth) led to the dominance of melanin-containing species with large, multicellular conidia both in the topsoil of all profiles and in the deep playa layers. At the same time, in the middle depths, mesic group of microfungi, *Penicillium* spp. producing light-colored small one-celled conidia mostly prevailed. In that way, the above vertical variations resemble differences in the composition of topsoil microfungal communities found between the sites located in the Negev desert and in the Mediterranean region of Israel. At the crusted sandy and playa habitats, density of microfungal isolates sharply decreased with depth. A depth-wise decline in isolate density, although not as sharp, was found also in the undercanopy sandy profiles, while in the bare profiles, this parameter did not substantially vary. The characteristics of microfungal communities showed relationship of different strength and direction with organic matter content, pH, and electrical conductivity. Substantial increase in isolate densities, an indirect indicator of the increase of fungal biomass, at the undercanopy habitats with higher substrate stability raising the accumulation of organic matter, highlights the role of shrubs as 'islands of fertility' for microfungal communities in arid lands.

PS - poster session

**Taxonomic composition of microfungal communities in organic layer under Japanese knotweed**

**Karolina Oszust**<sup>1</sup>, Magdalena Frąc<sup>1</sup>, Anna M. Stefanowicz<sup>2</sup>

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Invasive plants are recognized as a major threat to biodiversity. Although they are well-established in natural areas, the supposed negative impacts upon ecosystems, including soil microbial communities, have so far been poorly investigated. We focused on the Japanese knotweed (*Reynoutria japonica*), a widespread invasive species in Europe and North America, that produces massive amount of biomass, influencing the quantity and quality of soil organic matter. Fungi are well described to perform turnover of dead organic matter in soil, and their diversity might be characteristic for Japanese knotweed. Therefore, the goal of this work was to evaluate the core fungal microbiom taxonomic composition in organic layer under this species.

We considered five patches of *R. japonica* on fallows and riverside areas. The patches were characterized by > 90% *R. japonica* coverage. Organic layer samples were collected in autumn 2017. The isolation of the community DNA was performed using FastDNA SPIN Kit for Faeces® kit. It was followed by the barcoded Internal Transcribed Spacers (ITS) next generation sequencing. The MiSeq 2000 platform was applied to sequence the DNA of microfungal community. FUNGuild was used to taxonomically parse fungal operational taxonomic units (OTUs) by ecological guild. The most abundant and characteristic for all five tested sites were representatives of Ascomycota: order Dothideomycetes: *Phoma schachtii*, Pleosporales spp., Dothideomycetes spp.; order: Leotiomycetes: *Cadophora luteo-olivacea*; and order Sordariomycetes: *Trichoderma martiale*, *Sarocladium strictum*, *Fusarium sporotrichioides*, *Myrmecridium montseguinum* and *Discosia pseudoartocreas*.

This research was supported by National Science Centre (Poland), project No.: DEC-2016/23/B/NZ8/00564.

PS - poster session

**Changes of fungal diversity in soils amended with biofertilizers**

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The increase of the agricultural product demand (connected to the population increase) has led to an increase of the fertilizer used to improve the nitrogen content and the level of the soil quality. The excessive use of fertilizer can lead to groundwater pollution and increased greenhouse gas emission. The aim of the study was to find a correct combination of three different biofertilizers (mineral fertilizers combined with beneficial microorganisms) on the abundance and diversity of soil fungal community. Analysing the relative abundance of each fungal class, a different influence of the biofertilizer on the abundance of the fungal community in both the field was observed. The diversity index recommended that the two fields are different only for the abundance of the individual in each presented class and the Shannon index (lower for heavy soil). We could have noticed that the many fungal classes, inside the light sandy soil, were affected by the analysed treatment than heavy soil.

This work was financed by The National Centre for Research and Development in frame of the project BIOSTRATEG, contract number BIOSTRATEG3/347464/5/NCBR/2017.



PS - poster session

### **Cladoniicolous fungi in the Carpathian Basin**

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*Cladonia* (Cladoniaceae) is a highly diverse lichen genus with about 500 species worldwide. Approximately 10% of the species are found in the Carpathian Basin.

*Cladonia* species are easy to recognise in the field on lowland sandy steppe areas with xerothermic vegetation, in mountain rocky grasslands or in woodland ecosystems. Lichen collections contain a huge amount of identified *Cladonia* material. Studying these materials helps to discover lichenicolous fungi, which are otherwise difficult to collect, since some of them do not cause conspicuous malformations or damages on the host thallus.

So far 128 obligately lichenicolous species are known from *Cladonia* species.

*Cladonia* thalli are rich in lichen secondary metabolites as our investigations confirmed also in Hungarian specimens. These might support or prevent the colonization of lichenicolous fungi. As we observed during our experiments studying UV screening cortical pigments, a *Didymocyrtis* species was the most abundant lichenicolous fungus parasite on *Cladonia* samples causing decolourisations both on the margin and in the middle of the lobes and thalli.

The most frequent cladoniicolous species will be presented together with their hosts and habitats. Comparison of lichenicolous fungi of host species living among different environmental conditions may increase our knowledge on the indicator role of these fungi.



PS - poster session

**The diversity of fungi associated with *Quercus rubra* (L.): the case of 100-year-old alley in the city center**

Mikołaj Matela, Anna Frymark-Szymkowiak, Magdalena Kulczyk-Skrzeszewska, Barbara Kieliszewska-Rokicka  
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Trees are significant components of urban environment, however exposition for many stress factors bring down their resistance from diseases. Trees attacked by pathogens are sources of infection of healthy trees. Therefore an assessment of the presence and extent of the wood decay process and identification of the fungi responsible for decay can improve the management of urban woody plants. The present work contains mycological expertise concerning red oaks (*Quercus rubra* L.), fast growing large tree with great decorative qualities, native of North America, introduced to Poland in 19th century. We investigated trees growing in the form of an alley in the center of Bydgoszcz, Poland. The alley was built in 1903 and covered about 100 trees. The objectives of this study were: 1) to assess the frequency of fruiting bodies of wood decay fungi in the red oaks trees and evaluate the risk of the old alley due to fungal infection, 2) to determine mycorrhizal status of fine roots of the oaks and to find the relationship between the health conditions of the oak trunk and the abundance and variability of mycorrhizal fungi communities. The study material comprised 91 trees. Five species of commonly occurring decay fungi were identified on the basis of fruiting bodies on trunks of 18 trees. The most frequent species were *Fomitoporia robusta* (= *Phellinus robusta*) and *Daedalea quercina*. In addition, the occurrence of *Laetiporus sulphureus*, *Pholiota squarrosa*, and *Schizophyllum commune* was registered. Tree roots were colonized by ectomycorrhizal fungi and, in a small percentage, by arbuscular mycorrhizal fungi regardless of the infection of tree trunks by wood decay fungi.

PS - poster session

**Data collecting on distribution of fungi in Latvia by communication with general public**

Inita Daniele, **Diana Meiere**

*Latvian Museum of Natural History, Riga, Latvia*

Mycological knowledge on fungus diversity and distribution of species in Latvia is very scarce. The Latvian Museum of Natural History makes a collection of local mycoflora and works to raise public awareness on biological diversity and conservation in general, and mushrooms in particular. There are very few professional mycologists in Latvia, and currently we are looking for possibilities to find new ways of getting additional information encouraging the general public to get interested in mushrooms and help to fill in some gaps in data on fungi in Latvia.

The Museum is the main place where people can get their mushrooms identified. Samples brought for identification, especially during our annual mushroom exhibition, sometimes prove to be rare or even new species to Latvia.

Several NGOs are working in connection with nature diversity. Members of Latvian Mycological Society – mostly amateurs – are very helpful to professionals. Society organizes mushroom forays and announces the Fungus of the Year. For example, Fungus of the year 2019 is *Sarcodontia crocea*, and the first report about this fungus had already been received at the beginning of the year. Mycological society has also its Facebook page and number of followers is growing.

Another source of valuable information on the distribution of fungal species is a nature observations portal [dabasdati.lv](http://dabasdati.lv) where everyone is welcome to share their observations in nature – report on wild plants, animals and fungi. Reports on many rare fungi (*Lentinus tigrinus*, *Erythricium aurantiacum* etc.) come from that site.

Data acquired from general public is invaluable for mycologists, bearing in mind that there is just a few full-time professional mycologists and scarce resources for research of fungal diversity and distribution in Latvia.

PS - poster session

**Macrofungi of Shar Planina the largest mountain range in the Central Balkan**

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Shar Planina as a momentous Central Balkan mountain range has diverse geology, geomorphology and many glacial lakes, with continental, mountainous and alpine climate presents a mountain with very rich biodiversity, very important from the conservation point of view (Melovski et al. 2013). According to the published papers and data base information a total number of 497 fungi species are known for Shar Planina (Rusevska, Karadelev 2017).

In more than two decades of field activities on this mountain, in a variety of habitats and substrates a lot of material was collected. A significant number of exsiccates were a subject of taxonomical analyses in the last year, which enlarged the previous known list of fungi on Shar Planina.

According to the present data a total number of 761 taxa are listed for Shar Planina. Most of them belong to the phylum Basidiomycota, in total 646 of which 31 belong to the gasteroid fungi, while the rest to Ascomycota (108) and only seven to Myxomycota. According to the substrate 358 species are terricolous, 403 lignicolous and 19 taxa are hypogeous. Among the identified species, 32 are part of the Macedonian Red List of Fungi, as critically endangered are: *Discina parma*, *Galerina jaapii*, *G. sphagnorum* and *Galerina tibiicystis*, while endangered are *Amanita caesarea*, *Helvella atra*, *Omphalina grossula* and *Pleurotus eryngii*.

The authors express their gratitude to the Biology Students' Research Society and Macedonian Ecological Society for arranging many field trips on various localities on Shar Planina resulting in valuable collections. The research of the fungi diversity also received support from Rufford foundation in the frame of the project: Fungi of Kosovo: Establishing Fungi Checklist and Preliminary Red List, and Proposing Important Fungal Areas.

PS - poster session

**Fungi colonizing different parts of tomato plant (*Solanum lycopersicum*) in Slovakia**

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A diverse group of organisms, including fungi, colonizes leaves and root of tomato plants (*Solanum lycopersicum*). The aim of the study was to isolate, determine, and identify mycobiota inhabiting leaves and root of tomato, based on morphological characteristics of fungi. Tomato plants were sampled at the end of the 2018 growing season in Slovakia. A total more than 500 tomato plants were analysed from 35 different commercial cultivation sites.

Symptomatic leaves of *S. lycopersicum* were selected to evaluate fungal diversity. Fungi on tomato leaves were identified using morphological traits. The most abundant fungal species were *Septoria lycopersici*, *Alternaria solani*, *A. alternata*, *Phoma* sp., *Stemphylium* sp., *Cladosporium* sp., *Passalora fulva* and *Phytophthora infestans*.

A total of 2.384 isolates of fungi were identified from tomato root. The isolation and morphological determination of the fungi were performed on PDA and Czapek's nutrient media. Isolates derived from diseased tomato root were grown in pure culture and identified. The majority of fungi consisted of Deuteromycetes (95.7%) followed by Ascomycetes (4.1%). The main fungal taxa observed in tomato root in vitro were *Colletotrichum* (59.5%), *Fusarium* (10.4%), *Verticillium* (2.1%) and *Rhizoctonia* (1.1%). Several species from genus *Colletotrichum* and *Fusarium oxysporum* are considered primary components of the common root complex in Slovakia. The results of this study will help Slovak farmers to focus on appropriate management of emerging and recurrent fungal diseases of tomato.

This work was supported by the Slovak Research and Development Agency under the contract No. APVV-17-0150.



PS - poster session

**Soil contamination as a key factor influencing fungal diversity**

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Fungal communities are crucial for soil functioning and cycling of organic and inorganic matter. Although bioremediation studies aiming for characterization soil fungal communities from contaminated sites were conducted, usually, due to methodological limitations, these were focusing on selected groups of fungi, e.g. mycorrhizal ones. Nowadays, along with the development of modern techniques, such as next generation sequencing, more complex studies of soil fungal communities became possible. In the presented study, we aimed to verify whether soil contamination with metals and polycyclic aromatic hydrocarbons (PAHs) influences the structure and diversity of fungal communities. For the analysis, 100 soil samples from polluted and control (pristine) remote regions in Poland were collected. All samples were characterized in terms of physical and chemical parameters (including: humidity, pH, PAH and metals content). Insights into taxonomic composition of fungal communities in analysed sampling sites was assessed using sequencing of the PCR amplicons of ITS2 region using Illumina MiSeq platform. Finally, obtained exact amplicon sequences variants (ASVs) were assigned to more than 1000 known genera using Unite database as a reference. It was shown, that representatives of the genus *Mortierella* were the most abundant group of soil fungi and they were present in all tested soil samples. The common feature of PAHs-polluted soils was significant dominance of Venturiales representatives. Fungal communities in metal-polluted soils seem to be less distinctive. Our results suggest that type of soil contamination is one of the critical factors shaping soil fungal community.

The study was supported by the National Science Centre, Poland, under grant no. 2017/25/B/NZ8/00473

PS - poster session

**Fungal endophytes in common wheat (*Triticum aestivum* L.) cultivated in Poland**

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The group of fungi occupying internal parts of the plant without leading any disease syndromes in their host are termed as endophytes and have substantial impact on plants. Endophytes are frequent observed in nature and demonstrate positive (mutualism) or none (commensal) effect on host plant, but also due to combine lifestyles, some of them are latent pathogen and dormant saprobes. Symbiotic endophytes generate beneficial effects: can blocked pathogen development, promote host growth and increase host defence against abiotic stresses.

The aim of our study was to explore the mycobiome structure in endosphere of winter (Rospuda, Rusalka, Bombona, Kandela, Arabella) and spring (Bamberka, Ostroga, Arkadia, Legenda, Euforia) wheat cultivars grown under controlled conditions and to evaluate fungal distribution in four plant organs: leaves, stems, seeds and roots. Identification of endogenous fungi was performed by DNA sequencing of the internal transcribed spacer region (ITS) region, small-subunit (SSU) or large-subunit (LSU) nrRNA and protein coding markers: beta tubulin (tub2), translation elongation factor 1-alpha (tef1) and the largest subunit of RNA polymerase (RBP1). Altogether 225 fungal isolates were obtained. In all examined cultivars and organs *Sarocladium* spp. and *Penicillium* spp. were presented. The differences in fungal species composition between host organs were noticed. *Clonostachys* spp., *Engyodontium* spp. and *Aspergillus* spp. were identified only in seeds, while *Trichoderma* spp. and *Fusarium* spp. were observed mostly in roots. Furthermore, *Cladosporium* sp., *Acremonium* sp., *Phlebia* sp., *Marasmius* spp. and *Geomyces* spp. were detected.

This study was supported by the National Science Centre in Poland Grant No 2017/27/B/NZ9/01591

PS - poster session

**Hidden from sight - mycorrhizal fungal diversity in South African soils**

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Distinct types of these mycorrhizal associations are recognised based on the fungi and plant species involved as well as the symbiotic structures formed within or around plant roots. In South Africa ectomycorrhizal (ECM) fungi have generally been introduced with exotic commercial plantation trees, while the indigenous vegetation is mainly arbuscular mycorrhizal (AM) with the exception of the members of the Ericales which are ericoid mycorrhizal (ErM) and orchids which harbor a unique orchid mycorrhizal (OM) association. Using both culture dependent and independent approaches ECM diversity below ground is far greater than previously recognized from above ground fruiting bodies. AM fungal diversity in various environments is dominated by species in the *Ambispora*, *Glomus* and *Paraglomus* genera with a rarer components represented by species in the *Redeckera*, *Archaeospora* and *Geosiphon* genera. ErM fungal root associates were predominantly from the Ascomycota, although Basidiomycota and Glomeromycota (AM fungi) were also detected. Disa roots were colonized by the OM *Tulasnella* species such as *T. colaspora*, while Eulophia roots were colonized by *Psathyrella condolleana*. Although common in all ecosystems mycorrhizal biodiversity in South African soils and plants have received little attention. Research suggests a greater diversity of mycorrhizal fungi and land-use practices may negatively impact this unknown diversity an associated ecosystem services.



PS - poster session

**Microfungi (incl. larger Ascomycota) of the Białowieża Forest – synthesis result after 130 years of investigations**

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The Białowieża Forest is well known place throughout Europe. Forests in this area have been around for millennia. They are the only relict of former extensive Boreal-Nemoral forest zone in Europe. More than 100 years old, natural and most preserved stands show high richness of plants species, varied age and layers structure, as well as abundance of dead wood lying down in the forest floor. Ecological processes occur naturally here and stimulate self-renewal of the forest without human intervention. The Białowieża National Park was created in the most valuable and preserved areas.

At the same time, they support appearing of a rich biota of fungi. Białowieża Forest is recognized both in Poland and in Europe as one of the most important fungal refuges. The history of mycological research is over 130 years long. The authors of the report undertook a challenge of preparing a critical list of organisms classified as microfungi. The list is quite long with about 1,474 species. They belong to true fungi (1293 species) and two groups of fungus-like organisms – protozoan fungi (113) and chromistan fungi (68). Among the true fungi the most numerous is Ascomycota phylum (1073), followed by Basidiomycota (179), Chytridiomycota (3) and Zygomycota (38). The work provides a full list of all known fungal localities, host organisms, substrates, habitats, plant communities, together with their time of the collection.

Most fungi were collected on live plants, their dead remains, forest litter, and insects. The research was carried out (apart from forest communities) also on non-forest habitats. However, no detailed studies were conducted in this direction. The number of microfungi that could be found in Białowieża Forest is certainly much higher than it has been shown so far.





PS - poster session

**ITS diversity of the soil mycobiota in a Mediterranean forest ecosystem**

**Paulo Oliveira<sup>1</sup>**, Beatriz Lourenço<sup>1</sup>, Teresa Freire<sup>1</sup>, Marco Guerreiro<sup>2</sup>, Derek Peršoh<sup>2</sup>

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Approaching soil mycobiota diversity through metagenomics has brought a dazzling richness of data, confirming the previously suspected high  $\alpha$ -diversity, even in the tiniest of samples, the high number of undescribed taxa, and the patchiness of community composition. However, in the coverage of Mediterranean forest ecosystems, the soil metagenomics literature for fungi is rather thin. The present study is a proof-of-concept effort, based on a novel DNA extraction method, to explore the contrasts between a pair of small adjacent plots in a forested Mediterranean area where *Quercus suber* L. is the dominant tree species. A set of 26 sample points forming a grid-type lattice were collected and processed within a short time period on December 2017. For each point, two soil depths were sampled, one just below the soil litter and the other at least 20 cm deep, marked by a sharp decrease of organic matter. Most studies so far are designed only with the top layer. It was hypothesized that, in spite of the patchiness, there would be a preferential clustering between samples of the same plot, and between those of the same depth. Both hypotheses are strongly supported by the data, only less so for the plots contrasts in the deep samples, due to the very high  $\beta$ -diversity at this depth. Some OTUs were localized topographically, while others were ubiquitous at this (small) scale. Such ubiquity would probably be missed by taking just one (or a few) samples per plot.

PS - poster session

**Macrofungi Diversity in the Middle East: Survey and Identification of Wild Mushrooms in Lebanon**

**Nadine Modad**

*American University of Beirut, Beirut, Liban*

Few studies have been conducted on the identification of macrofungi in the Middle East. In this region, mushrooms present little economic importance except for a few species such as some agarics, which are traditionally harvested from the wild. The objectives of this study consisted of surveying, collecting, identification and preservation of wild mushrooms. The methodology consisted of: 1) survey trips and field observations, and 2) laboratory work including macroscopic and microscopic records and preservation of specimens. The habitats visited included oak and pine forests, mixed woods, cedar reserves, meadows, riversides, gardens, orchards and rare habitats. There is generally a large flush of growth in the autumn with a minor one in the spring. The most dominant species found were the Agarics, Lepiotas, Tricholomas, Russulas and Boletes. Because very few meadows are present in Lebanon, the most promising stations for mushroom appearance were found to be coniferous and cedars forests, pinewoods, and oak groves. The total number of specimens collected was five hundred and two specimens out of which three hundred and sixty eight species were fully studied and identified. These included thirty-two species belonging to the Ascomycetes and three hundred and thirty six species belonging to the Basidiomycetes. Among the latter, four species were coral fungi, one chanterelle, two jelly fungi, two thelephoras, ten teeth fungi, seven crust fungi, eighteen bolete species, twenty two polypore species, twenty two gasteromycetes and two hundred and forty eight species of the gill fungi. Compared to many arid and semi-arid countries in the Middle East, the richness of humic substances in multiple areas of Lebanon favors the proliferation of hundreds of species of mushrooms.

PS - poster session

**A widely applicable approach to fungal communities succession**

**Paulo Oliveira**, Cláudia Lopes, Gonçalo Isidoro, Rodrigo Teles, Carlos Vila-Viçosa  
*CIBIO - Research Centre in Biodiversity and Genetic Resources, Évora, Portugal*

It is well-known that, as vegetation changes in an evolving habitat, the mycoenose will necessarily change. Thus the ecological niche may, at least in some fungal species, be circumscribed by the successional stages they thrive in. However, in contrast to plant communities succession, the fungal counterpart remains difficult to approach, and the available results are mainly from wood decaying communities or community sequences observed after major disturbances. Nevertheless, the interdependence between the vegetation and mycobiota in each ecosystem offers an opportunity for other settings. Specifically, an approach guided by a well-characterized set of plant succession stages may uncover previously unsuspected successional correlates in the mycocoenoses. In this communication we present the results of four years of studying the occurrence of macrofungi in two adjacent cork oak plots with a sharp successional demarcation. Recurring contrasts in the saprotroph and biotroph fungal assemblies were recorded, and a strategy for the analysis of these contrasts is proposed, to highlight the information gained on the ecology of at least some of the species. Preliminary results from a soil metagenomics study covering these two plots will also be presented. Finally, the presentation will suggest ways to replicate the proposed approach, and its perceived relevance with regard to processes such as global change and land use.

PS - poster session

### **Fungal diversity of Jagiellonian tapestries**

**Lukasz Istel**<sup>1</sup>, Magdalena Dyda<sup>2,3</sup>, Paulina Drabik<sup>2</sup>, Magdalena Ozga<sup>4</sup>, Jerzy Holc<sup>4</sup>, Karolina Szlęka<sup>4</sup>

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Dated to the middle of 16th century, Jagiellonian tapestries are one of the most valuable and interesting textile collections in the world. Originally the collection consisted of about 160 textiles of different kinds and sizes including figural tapestries and verdure but only 136 of them survived up to nowadays. Currently, they are kept in the museum of Wawel Castle in Cracow, where they are regularly vacuumed.

The biggest risk to the whole collection is biodeterioration by enzymes produced by filamentous fungi, that may cause a decrease of strength properties by decreasing the level of fibre polymerisation. Additionally, fungal enzymes can decolorise textiles by reacting with pigments in fibres.

The aim of the present study was to determine the diversity of mycobiota of tapestries and control the effectiveness of maintained treatment to prevent biodeteriorative impact of fungi.

Samples were collected using sterile cotton swabs and sterile cellulose fragments. Different agar media were used for isolation of bacteria and fungi. Plates were incubated for 2 weeks and the fungal taxa were identified using classical morphology-based identification to genus level.

The most abundant fungi taxa were frequent airborne genus e.g.: *Penicillium*, *Cladosporium*. Biodeteriorative fungi occasionally were also noted e.g. *Trichoderma*, and human pathogens e.g. *Blastomyces* as well.

PS - poster session

**Fungal richness in Acute Oak Decline infected *Quercus robur* L. in Latvia**

**Lauma Brūna**, Natālija Burņeviča, Tālis Gaitnieks  
*Latvian State Forest Research Institute "Silava", Salaspils, Latvia*

One of the recently discovered disease in Europe is Acute Oak Decline (AOD) that cause oak dieback. Due to this disease tree can die in 3 to 5 years. First records of AOD are from United Kingdom but it keeps spreading through Europe. It was discovered in Latvia in 2017.

The main disease agents are considered to be bacteria *Brenneria goodwinii* and *Gibbsiella quercinecans* but there may be other factors that affect spread and distribution of AOD and determine tree resistance against bacteria. The aim of the study was to assess pathogenic fungi richness in *Q. robur* infected by AOD in Kurzeme region in Latvia.

Wood samples were taken from trees with disease symptoms and healthy looking oak at root collar and necrotic stem lesions. 33 samples from diseased trees and 33 samples from healthy trees were taken at root collar, as well as 33 wood samples from stem lesions.

In total, 27 fungal species were isolated. Significant difference between fungal species richness in diseased and healthy oaks was not observed. Pathogenic fungi were found in both healthy and infected trees. From stem lesions 9 fungal species were isolated. Some of isolated fungi may cause faster oak decline. The study is ongoing and more data will be observed.

PS - poster session

**Innovative silvicultural treatments to enhance soil biodiversity in artificial black pine stands: effect on macrofungal diversity**

Claudia Perini, Debora Barbato, Elena Salerni  
*University of Siena, Siena, Italy*

Thinning, as a man-induced disturbance of forest ecosystems, may affect biodiversity and other related ecosystem functions. In this context, a multidisciplinary EU-Life project (SelPiBioLife) was established in 2014 aiming at evaluating the application of an innovative forest management technique in two *Pinus nigra* plantations areas of the Apennines (Tuscany, Italy). Respect to traditional management techniques, innovative thinning is based on the selection of the most productive trees and the regulation of sunlight through the modification of forest cover. This treatment was applied to test the effect on stand stability, tree growth rates and various components of soil biodiversity (flora; fungi as microorganisms, ectomycorrhizae and mushrooms; mesofauna; nematods; microarthropods; bacteria). Here we present the results regarding the Pratomagno forest mushrooms above ground one year before (2014) and 4 years after (2015-2018) the silvicultural treatments (traditional, selective and no-thinning). Over the whole study period, 191 fungal species, 14064 carpophores, with a fresh and dry weight of 103.26 kg and 10 kg respectively, were observed. After the expected decrease in the first year post-thinning, total species richness did not show a constant trend in its response to treatments effect. Concerning abundances, selective thinning showed the most evident fluctuations in short and mid-term. With regard to difference in species composition, the traditional treatment, after a first post-thinning stress, remains more or less stable and similar to the control; on the other hand, the selective thinning continues to progressively increase its dissimilarity with respect to the control, showing a different guild of species in the last post-thinning year.



PS - poster session

**MycoIndoor: citizen science to unravel the mycobiomes of Norwegian houses**

**Pedro M. Martin-Sanchez**<sup>1</sup>, Eva Lena F. Estensmo<sup>1</sup>, Maria Nunez<sup>2</sup>, Johan Mattsson<sup>2</sup>, Ingeborg B. Engh<sup>2</sup>, Inger Skrede<sup>1</sup>, Håvard Kauserud<sup>1</sup>

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<sup>2</sup>*Mycoteam AS, Oslo, Norway*

Fungi have ubiquitous distribution in natural ecosystems as well as in built environments, where people spend major parts of their lifetime. When moisture is present, fungal growth can cause important problems such as material deterioration and adverse effects on the occupant's health. Previous studies have stated that geography and climate are the most important determinants of indoor mycobiomes, with the outdoor air as the key source of spores. Norway possesses a marked climatic gradient that enables interesting microbiological studies on the built environment. However, the mycobiomes of Norwegian buildings have not been explored yet.

MycoIndoor is a citizen science project aimed to improve the knowledge about the indoor mycobiomes of Norwegian houses by combining dust sampling with high-throughput amplicon sequencing (DNA metabarcoding). In May 2018, 291 volunteers participated in this study collecting and sending back three dust samples from their houses (outside, living room and bathroom) located across Norway. Dust mycobiomes were analyzed by DNA metabarcoding of ITS2 region and statistically compared with diverse climatic data and building characteristics through multivariate analyses. Results from this study will characterize the geographical variation of the indoor mycobiome in Norway and reveal the main environmental drivers at different scales.

PS - poster session

**Xylotrophic pathogens in orchards of the Middle Russian Upland: new hosts and new localities**

Sergey Volobuev

*Komarov Botanical Institute of the Russian Academy of Sciences, Saint Petersburg, Russia*

The territory of the Middle Russian Upland occupied approximately 480,000 km<sup>2</sup> is one of the key agricultural areas of European Russia. Human-transformed landscapes are presented by cities, fields of grain and vegetable crops, orchards, as well as urban and rural settlements with individual farms. The species diversity and distribution of macrofungi in such anthropogenic habitats as orchards and gardens has been underestimated here more than 60 years till now. The data on polypores and hydroid lignicolous fungi and their hosts from this area was listed by A.S. Bondartsev (1953), T.L. Nikolajeva (1961), and I.S. Popushoy (1971). The further target studies were initiated only in last decades mainly on the territory of forests adjacent to orchards. Main cultivated fruit trees in the Middle Russian Upland are apple-tree, pear, plum and cherry-tree. In total, 16 species of aphyllorphoroid fungi were registered as pathogens infecting living trees of *Malus*, *Pyrus*, and *Prunus*. Among them not only well-known and expected *Sarcodontia crocea* and *Phellinus pomaceus*, but also *Bjerkandera adusta*, *Chondrostereum purpureum*, *Coniophora puteana*, *Inocutis rheades*, *Oxyporus obducens*, *Radulomyces confluens*, *Trametes hirsuta*, *T. ochracea*, *T. pubescens* and some others are revealed. New localities of these species in orchards of the Belgorod, Kursk, Lipetsk, and Oryol regions, will be presented, and host spectrums of wood-inhabiting pathogens will be discussed.

The study was funded by the Grant of the President of the Russian Federation, project MK-3216.2019.11.





PS - poster session

**Letters (on fungi) from small islands - comparisons from around the world**

**Howard Fox**<sup>1</sup>, Maria Cullen<sup>2</sup>

<sup>1</sup>*National Botanic Gardens, Dublin, Ireland*

<sup>2</sup>*Barrow Herbarium, New Ross, Ireland*

Since 1987 a quest has been underway to explore the vast array of fungi, especially lichens, on small islands, beginning with Ireland and its islands. The aims have been taxonomic in the main, along with enhancement of the knowledge of species distributions on small islands around the world. These islands include Jamaica, Saint Lucia, New Caledonia, Tahiti, Moorea and Niue Island.

Alexander von Humboldt composed physical tableaux of species and created accessible infographics demonstrating the geographical distributions of plant communities. This type of work is considerably easier now and it lends itself to greater ecological understanding and effective conservation management. On high islands like Saint Lucia, maps of species distributions indicate patterns relating to altitude so habitat boundaries are identifiable. Contrasting lithologies such as coral limestone and ultramafic rock host distinct communities of lichens e.g. New Caledonia. Differences of corticolous, saxicolous and terricolous ratios between islands are due to a range of factors for example forest cover, humidity and wind exposure. Movement of host plants by humans from one part of the world to another can involve spread of epiphytic species over great distances e.g. *Cocos nucifera* and *Mangifera indica*.

Many small island scientific communities do not have considerable resources for the documentation of species or for conservation needs of indigenous fungal biodiversity. Appreciation of the challenges for small island dwellers across the world can be of use in knowing what specific knowledge and resources are required to establish new networked herbaria, to develop expertise and to build communities of local and international collaborators. In the end, we are all on a small, vulnerable island together.

PS - poster session

### Do tourists matter?

Magdalena Dyda<sup>1,2</sup>, **Lukasz Istel**<sup>3</sup>, Paulina Drabik<sup>1</sup>, Sławomir Korzeniowski<sup>4</sup>, Piotr Jarek<sup>4</sup>, Maria Kisiel-Jarek<sup>4</sup>

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Cracow is the top touristic city in Poland and becomes one of the most popular destinations in European tourism. Unfortunately, Cracow is one of the cities with the worst air quality in Europe. The Wawel Royal Castle is the central spot in the city and one of the most important museums in Poland (in 2017 almost 1.5 million tourists visited Castle). These two factors make the maintenance of historical heritage very hard. On the one hand, Castel staff need to make Museum tourists-friendly and, on the second hand, create a climate that keeps the collection safe from pollution - especially biological ones.

In present study In Air Pollution (IAP) were monitored, including a concentration of gaseous, particulate matter (PM) and microbiological aerosols. The main factor in room selection was tourist-trafficking. State Rooms, Royal Private Apartments, and conservation workrooms were studied. Bioaerosols samples were cultivated in room temperature for 2 weeks and identified using morphological characteristic to genus level.

The first hall, Castle entrance and some of the conservation workrooms had the highest concentration of PM, selected chemical compounds and bioaerosol. 33 different genera of filamentous fungi were isolated from the air samples, including biodeteriorative and human pathogen taxa.



PS - poster session

**Characterizing fungal microbiota of Romaine lettuce using Sanger and next-generation sequencing platforms**

**Danny Haelewaters**, Samira Fatemi, Makenna L. Houston, M. Catherine Aime  
*Purdue University, West Lafayette, USA*

Many outbreaks of human pathogenic bacteria, such as *E. coli*, are associated with Romaine lettuce, posing a threat for human health. We are characterizing fungal microbial communities inhabiting the phylloplane of this vegetable crop. We examined 63 Romaine lettuce heads acquired from grocery stores in Indiana, Illinois, Ohio, and Virginia. From each lettuce head a sample of 25 g of leaves was homogenized in 225 mL 100 mM phosphate buffer. Aliquots of 100  $\mu$ L were plated on YMA and PDA with antibiotics. After incubation at room temperature for 4–5 days, fungal colonies were sub-cultured until axenic. We isolated 348 strains from our lettuce samples. These strains were DNA barcoded with the internal transcribed spacer region of rDNA. We generated ITS sequences for 255 strains, representing 63 unique species of fungi on the lettuce phylloplane. Of these, several are undescribed species of *Cystofilobasidium*, *Holtermanniella*, *Rhodotorula*, *Sampaiozyma*, *Sporobolomyces*, and *Tilletiopsis*. One undescribed species of *Sporobolomyces* (Pucciniomycetes, Sporidiobolales) is the most commonly encountered species in our cultures. Next-generation sequencing was conducted on the same homogenates, resulting in 630 operational taxonomic units. We compared abundances at different taxonomic levels (classes, orders, genera) between treatments. The fungal abundance at all levels was highest in organic lettuces, closely followed by conventional lettuces. Fungal abundance in hydroponic samples was almost non-existent. Our results indicate that certain groups of fungi, like yeasts, in organic samples are replaced by others, such as molds, in conventional samples. The next step in our characterization will involve understanding of how some of these fungi interact with the human pathogenic *E. coli*.

PS - poster session

**Rhizosphere of wild raspberries as a source of beneficial fungi**

Michał Pylak, Karolina Oszust, Magdalena Frąć  
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It is hard to deny that modern agriculture, in particular organic farming, is focused on finding new solutions in plant protection. Fungal diseases in plants can cause serious yield losses. Beneficial microorganisms are mostly plant growth promoting bacteria and fungi. They are able to enhance nutrient uptake by plants, parasitize on plant pathogens, induce resistance of plants, or secrete hormone-like compounds. Due to the fact, that plant roots play a major role as a carbon source in soil, some microorganisms adapted to living in association with roots in the narrow region of soil that is directly influenced by root secretions called rhizosphere. It has been proven that beneficial microorganisms introduced to plants rhizosphere are able to modify natural root microbiome enhancing plant defense against pathogens.

During our research we have obtained 31 rhizosphere soil and root samples from wild raspberries from 10 locations in Lubelskie voivodeship (Poland). Fungi from soil and root samples were isolated and identified, afterwards they were checked for antagonisms against 4 fungal plant pathogens. From 161 fungal isolates 41 species were identified as potentially beneficial. Further conducted research showed that 36 isolates showed satisfactory results inhibiting growth of previously mentioned pathogens 23 of them belonged to *Trichoderma* genus, and 2 to *Mucor* genus. Despite the fact, that *Mucor* genus is commonly known for its pathogenic properties, newly conducted research shows that it has stimulating effect on growth of some plants, thus it might be considered as beneficial.

This work was financed by The National Centre for Research and Development in frame of the project BIOSTRATEG, contract number BIOSTRATEG3/344433/16/NCBR/2018.



PS - poster session

**Fungi as alien as their host? Microfungi associated with northern red oak (*Quercus rubra* L.) in Poland**

**Malgorzata Ruszkiewicz-Michalska**<sup>1</sup>, Beata Woziwoda<sup>2</sup>, Malgorzata Stasinska<sup>3</sup>, Sebastian Piskorski<sup>4</sup>, Zuzanna W. Botew<sup>4</sup>

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Species composition of fungi inhabiting leaves of northern red oak, a North American tree species introduced and cultivated in Europe, have been investigated in Poland. Forest-planted trees as well as wild growing individuals were surveyed in western and central Poland. The study yielded 19 fungal species belonging to Ascomycota and observed exclusively in anamorphic state. Four of them (*Chaetomella acutiseta*, *Cladosporium fumagineum*, *Gnomoniopsis clavulata*, *Marssonina quercina*) have not been reported in Europe until now, and the other four species (*Phloeospora associata*, *Phyllosticta quercus-illicis*, *Seimatosporium glandigenum*, *Septoria querceti*) have been detected in Poland for the first time. Eight of the fungal species have not been reported worldwide as hosted by *Quercus rubra* although most of them are specialized to inhabit other *Quercus* species. None of microfungi species found during this study caused any serious disease symptoms, except for *Phloeospora associata*.

PS - poster session

***Dothistroma septosporum* mating types observed in Slovakia**

**Emília Ondrušková**

*Slovak Academy of Sciences, Institute of Forest Ecology, Department of Plant Pathology and Mycology, Nitra, Slovakia*

Identifying the extent that intraspecific diversity in mating types contributes to the presence of fungal pathogens across differing forest systems can aid conservation and management measures. Naturally regenerated or planted (urban greenery, forest plantation, arboretum-, and forest nursery) *Pinus* localities were sampled during 2013- 2019 to assess the deterioration of pine tree vitality in Slovakia. Symptoms included that the needles were green on the base and necrotic at the tip, with several red bands around the needles that were observed mainly on necrotic parts. Dark brown to black erumpent fruiting body (acervulus) presences could occur in the middle of the red bands.

DNA from 406 cultures and 32 symptomatic needles was successfully extracted and used for species and mating type determination. All samples were identified as *Dothistroma septosporum* (Dorog.) M. Morelet, which causes *Dothistroma* needle blight (DNB) -one of the most economically important diseases of *Pinus* sp. in the world. Twelve *Pinus* species were identified as hosts of *D. septosporum*. Mating type MAT 1-1-1- was identified in 197 samples (in 184 pure cultures), MAT 1-2 was detected in 254 samples (in 241 pure cultures). Both mating types simultaneously were identified in 8 needles and 5 culture samples. Out of 61 verified localities, in 29 of them the existence of both mating types was shown, and their presence was determined as a prerequisite for teleomorphic stage development. Our results demonstrate that, with respect to stand type, the mating types of DNB are equally distributed and not a source of reproductive limitation.

This work was supported by funding from the Slovak Research Grant Agency VEGA project no 2/0077/18 and from The Slovak Research and Development Agency APVV project no. SK-FR-0025.

FUNGI IN PRIMAEEVAL FORESTS AND OTHER NATURAL  
HABITATS

OS - oral session

**Plot-based macrofungi monitoring in taiga zone of West Siberia and sampling event data publishing**

**Nina Filippova**  
*Yugra State University*

The protocols for sampling macrofungi using fixed-sized plots have begun to converge in recent years (O'Dell et al. 2004) thus making possible comparison of different project results and allowing extrapolation of data. On the other side, development of integrated biodiversity information facilities has opened the possibility of standardized data storage and open access to the accumulated quantitative data.

Following the standard plot-based protocol (10.17504/protocols.io.z2xf8fn) for sampling macrofungi, we initiated the long-term monitoring program in major ecosystem types in middle taiga zone of West Siberia, nearby Khanty-Mansiysk. The goal is to describe the quantitative and qualitative structure and spatial variation of the community of macromycetes, to follow its dynamics seasonally and inter-annually. A series of plots was established in a raised bog ecosystem and in different forest types following after-logging and paludification successions (about 560 micro-plots totaling 1700 m<sup>2</sup>). The plots were visited weekly during seasons 2014-2018 and carpophores of different fungal taxa were counted in each micro-plot.

The sampling-event dataset type of Global Biodiversity Information Facility allows publication of data on quantitative community assessments. The assessments of macrofungi community are still absent or rare in GBIF, despite the importance of such data for ecological monitoring. Thus, the resulting monitoring data were organized as sampling-event datasets and published in GBIF to provide open access to the data (10.15468/e9g5ri; 10.15468/ge1hkl). The datasets include about 17000 plot-based observations (=sampling events) with corresponding about 6500 occurrence records. About 60% of plot-based observations contain zero records (absence data).



OS - oral session

**The occurrence and pathogenicity of *Geosmithia* fungal spp. associated with two cypress bark beetle *Phloeosinus* spp. in planted forests in Israel**

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Bark beetles are associated with fungal complexes, which vary in their ecological roles and specificity to their beetle and tree hosts. The relationship between beetles and their associated fungi ranges from obligatory to incidental, from mutualism to commensalism or antagonism. This study examined the diversity, distribution, vector affinity and pathogenicity of fungi isolated from two indigenous cypress bark beetles, *Phloeosinus armatus* and *P. bicolor* in Israel. The fungi were isolated from the beetle's body parts and from galleries, sampled from trap trees of *Cupressus sempervirens* exposed in several forest areas. A total of 273 fungal isolates were obtained, of these, 145, 19 and 114 were isolated from the beetles, larvae and galleries, respectively. Arbitrarily primed PCR was performed on genomic DNA of all 273 isolates using three primers (GACAC)<sub>3</sub>, (GACA)<sub>4</sub> and (CAG)<sub>5</sub> which delineated the fungal isolates into ten different groups. Multilocus sequence analysis was further performed using ITS 1 and 4,  $\beta$ -tubulin and the elongation factor genes on DNA of the ten representative isolates. The results revealed that all the beetle and gallery associated fungi belong to different *Geosmithia* spp., which was further corroborated with morphological characters. Pathogenicity assays were performed artificially on detached stems and on two-year-old cypress saplings under natural conditions in an outdoor net house. All isolates were pathogenic on detached stems but only one isolate exhibited a high degree of pathogenicity in cypress saplings. This study suggests a long and stable association between both *Phloeosinus* spp. and *Geosmithia* spp., and a possible role of certain associated fungal species as pathogens or endophytes towards this cypress tree.

OS - oral session

**Combined meta'omics reveal links among fungal community composition, gene expression, and chemical changes in decomposing leaf litter**

**Marco Alexandre Guerreiro**<sup>1</sup>, Stephan Kambach<sup>2</sup>, Raphael Stoll<sup>3</sup>, Andreas Brachmann<sup>4</sup>, Dominik Begerow<sup>1</sup>, Derek Peršoh<sup>1</sup>

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Decomposition of plant litter is essential for nutrient cycling and a key process for ecosystem functioning. Fungi are considered to be the main decomposers of leaf litter in forest ecosystems. They synthesize and secrete enzymes that change the chemical composition of the litter, and thus represent a major effect of the fungal community. However, fungal community composition and their metabolic activity have been rarely analyzed together, and so far never in combination with litter chemistry.

We characterized the chemical composition of autumn leaves of European beech (*Fagus sylvatica*) and the corresponding leaf litter after one year of decomposition by proton nuclear magnetic resonance (1H-NMR) spectroscopy. The composition and transcriptional activity of fungal communities was assessed by high-throughput Illumina sequencing in the same litter samples. These analyses were highly replicated across 14 different forest plots and cover three distant regions in Germany.

We were able to successfully distinguish freshly fallen leaves from one-year-old litter with respect to their chemical composition. Leaves were chemically more distinct among regions than one-year-old litter. Fungal communities were locally structured, however, functionally redundant among regions, i.e. expressing genes coding for similar litter-degrading enzymes. We identified changes in the abundance of putative chemical compounds between freshly fallen autumn leaves and one-year-old that correlated to the transcription level of litter-degrading enzymes. Transcription patterns were also correlated with the abundance of certain fungal species. Overall, we provide strong evidence of a dynamic interaction between substrate chemistry, expression of enzyme coding genes, and fungal community structure in nature.

OS - oral session

**Climate structures belowground fungal communities in semi-natural grasslands**

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Soil fungi are key players in terrestrial ecosystems and important drivers of long-term sequestration of carbon (C) in boreal soils. Experimental studies have shown that fungal growth is sensitive to changes in temperature and precipitation, yet we have limited knowledge about how climate change will affect fungal communities in boreal grasslands. In this study, we take advantage of steep climatic gradients in western Norway to assess the effects of variation in temperature and precipitation regimes on fungal community composition, activity and biomass in semi-natural grasslands. We show that while communities of fungi closely associated with plant roots are structured by both temperature and precipitation, soil-dwelling fungi only respond to changes in temperature. Soils in drier climates possess higher fungal biomass and soil C compared to wetter soils, which may be explained by higher relative abundances of recalcitrant melanized root endophytes in dry soils and climate-dependent decomposition processes. The predicted warmer and wetter climate in boreal areas may lead to lower soil C stocks in boreal grasslands, which ultimately may affect the global C balance.

OS - oral session

**Inside Oak – fungal community structure in the heartwood of oak trunks**

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Hollows and cavities are an important micro-habitat of old trees which are utilised by a diverse and often dependent set of other organisms. The gradual development of cavities by successional assemblages of wood-decay fungi leads to a functionally diverse habitat in both physical and chemical characteristics of the cavity.

In the European oaks *Quercus robur/petraea*, the primary colonisers of the heartwood appear to be a cosmopolitan group of basidiomycetes with a Holarctic distribution. However, few studies have systematically investigated the communities of fungi in oak heartwood, most of the available information being derived from sporocarp presence which may only be indicative of the actual community.

In this study we characterise the oak heart-rot community in *Q. robur/petraea* with a combined approach of traditional culture and molecular methods. Wood cores were extracted from undecayed stems and from the inside of cavities. Wood chips were removed from cores at 1 cm intervals and cultured on agar medium. DNA was extracted from mycelium and sequenced for identification using the ribosomal ITS universal barcode for fungi. Where no mycelium grew from a wood chip, but decay was apparent, DNA was extracted directly from the wood.

Although this project is ongoing, preliminary results indicate, as anticipated, that the fungal community is dominated by a small number of oak-specialist basidiomycetes. We have also found a high diversity of ascomycete taxa in relatively undecayed wood whose ecological function within the trunk is poorly understood.

Given the dominance of *Quercus* spp. in many wood-pasture and forest biomes, in the Northern Hemisphere, cavity creation and nutrient-cycling of heartwood may be concentrated in a disproportionately small number of oak specialists.

OS - oral session

**Low fungal host specificity of epiphytic orchids in a Neotropical canopy**

**Christoffer Bugge Harder**

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Members of the large Orchidaceae plant family are noteworthy for their complex mycorrhizal (mycoheterotrophic) relationship with certain orchid mycorrhizal fungal hosts (OMFs). While the mycorrhizal ecology of many terrestrial species has been studied extensively, less is known about neotropical epiphytes, even though these environments harbour the highest orchid species diversity. As all orchids depend on OMFs during germination and early development, the accessibility of suitably compatible OMF partners has frequently been suggested as an important constraint for orchid distribution.

We carried out a study on epiphytic orchids in the canopy of the ericoid *Saurauia montana* (host tree) in a lower montane rainforest in Costa Rica over four years, taking samples of orchid roots as well as from the epiphytic substrates (subdivided in live moss, dead moss, and bark). We sampled orchid roots and associated substrates from >300 points on 39 trees along a 250 m transect, and sequenced the ITS2 from substrate samples with Illumina, and from orchid roots with both Illumina and Sanger.

We identified 42 species of orchids from 551 orchid root samples from our study site, with over 50% representing 3 species (*Stelis hymenanthera*, *Oerstedella exasperata* and *Oncidium klotzschianum*). Sebaciniales and Atractiellales dominated among the OMF identified from roots. The OMFs found in the substrates showed considerable levels of patchy distribution and autocorrelation on distances below 100 centimeter; however, the orchid species showed few signs of specialisation and overall associated with multiple taxonomically divergent OMFs. Our data suggest that accessibility to compatible fungal symbionts may not be a limiting factor in the determination of the distribution of epiphytic orchids.

OS - oral session

**Diversity in buried wood - each running metre matters?**

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The difference between primeval and other types of forests can be expressed in number of fallen trees and their dimensions. As logs are one of the most important fungal habitats, their mycocoenosis in old stands will differ substantially from ones in managed forests. We carried the experiment in Białowieża Forest (BF), in which wood of four tree species (oak, hornbeam, spruce and pine) was exposed for seven years in three types of plant communities. We implemented NGS method to prepare metabarcoding data for 30 spruce logs buried in the ground. We revealed, that on the level of genera the fungi decomposing wood were not substantially differing from decomposers in forests planted by man. On the species level many common species were replaced in BF by rare or very rare fungi. Interestingly, the spruce logs buried in soil seem to be proper habitat even for numerous fungi known as mycorrhizal ones. Some of them were not recorded in the form of fruitbodies. In the experiment nearly all buried logs were covered by *Armillaria* rhizomorphs, regardless of the type of log and plant community, and nearly completely decomposed during seven years. In the molecular studies however *Armillaria* sequences were almost undetected. The comparison with data obtained from soil by Tedersoo et al (2014) reveals biocoenotical significance of the buried wood.

The study was supported by the National Science Centre, Poland, under grant no. 2016/23/B/NZ8/00897 and 2017/25/B/NZ8/00473

References

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PS - poster session

**Pathogenic fungi on *Prunus serotina* in selected localizations of north and south Poland - case study**

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The research was carried out between 2017 and 2018 from May to October, at selected locations in the Wigry National Park, within the city of Wrocław, as well as in the Lower Silesian Forests. The healthiness of *Prunus serotina* shrubs was assessed and mycological analysis of organs with disease symptoms was performed. In the area of Wigry National Park, leaf perforation (*Clasterosporium carpophilum*) and symptoms caused by fungi of the genus *Taphrina* were found. During the mycological analysis a total of 15 species were isolated. From the dying leaves 24.5% of the isolated colonies were *Fusarium avenaceum*, which may cause damage to the leaf blade. Leaves were also collected in which symptoms of leaf perforation (caused by *C. carpophilum*) were observed and as many as 66.6% of isolated colonies belonged to the *Arthrimum arundinis* species. From the dying shoots, a complex of fungi, mostly saprotrophic and weak pathogens was isolated, including: *Cladosporium herbarum* (22.54%) and *Alternaria alternata* (15.1%).

Also in Lower Silesia, the occurrence of *C. carpophilum* and *Taphrina* sp. was observed. Symptoms caused by both pathogens were more pronounced in the Lower Silesian Forests, compared to the city of Wrocław. No dying shoots were observed. The observed distortions occurred mainly on leaf blades, in the form of necrosis along the main nerve and spots on the edges of the leaves. In total, 16 taxa were isolated from the leaf surface, of which 27.95% was *A. alternata*, 13.04% *Aureobasidium pullulans* and 9.32% *Epicoccum nigrum*, which are weak pathogens or saprotrophs that build biofilms. Potentially pathogenic fungi were isolated individually: from the genus *Fusarium* (<4%), *Pestalotia* (<2.5%) and *Phoma* (<2%).

PS - poster session

**Spatial distribution of mycelial individuals in populations of two saprotrophic basidiomycetes *Mycetinis alliaceus* and *Gymnopus androsaceus***

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Although saprotrophic basidiomycetes play an important role in leaf litter decomposition little is known about spatial distribution and the size of individuals (genets) in this group of fungi. *Mycetinis alliaceus* and *Gymnopus androsaceus* are widespread in Northern Hemisphere and usually produce numerous sporocarps, which makes them a good model for population studies of litter-exploiting basidiomycetes. In this study, we analyzed three populations of *M. alliaceus* and two populations of *G. androsaceus* collected from four localities in the forests of the Republic of Serbia and the Republic of Montenegro. In order to determine distinct genets inter simple sequence repeats (ISSR) analysis was performed using five different primers ((GTG)<sub>5</sub>, (GACA)<sub>4</sub>, (GCC)<sub>5</sub>, (AG)<sub>8</sub>T and (AAG)<sub>6</sub>) separately. Majority of analyzed sporocarps showed unique ISSR profiles so each of them presents distinct individual (genet). Since all analyzed sporocarps were collected from a relatively small area and were found in close proximity it can be assumed that investigated populations consist of numerous and relatively small genets.



PS - poster session

**Relationship between environmental factors and macrofungal communities of different forest stands**

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In this study, we investigated macrofungal communities of different forest stands (beech / Douglas fir / Norway spruce and beech / Norway spruce, silver fir and beech) within several protected mountain areas in Serbia. During three years of sporocarp inventory on 5 permanent experimental plots, we identified 245 macrofungal species and classified them into three functional groups. Analysed environmental factors (precipitation, air humidity, air temperature, soil moisture), as well as the type of forest stand, had an impact on species richness and structure of macrofungal communities. Based on PLS regression, total species richness was influenced by air humidity and soil moisture, the richness of mycorrhizal fungi was mainly dependent on precipitation and air humidity, the richness of lignicolous fungi was influenced by air humidity, while a number of terricolous saprotrophs depended on soil moisture and to a lesser extent on air temperature. Canonical correspondence analysis was applied to investigate the distribution of recorded species within different habitats, based on environmental variables. Distribution of mycorrhizal species was highly influenced by precipitation, while soil moisture and air humidity had less impact. Air humidity and air temperature had the greatest influence on the distribution and composition of lignicolous species within different forest stands, with the majority of species preferring moderate values of studied factors. Variations in the composition of the terricolous saprotroph communities could not be attributed to the climate variables, but above all the differences between the forest stands.

PS - poster session

**Macrofungi on fallen oak trunks in the Białowieża Virgin Forest – ecological role of trunk parameters and surrounding vegetation**

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All groups of macrofungi were monitored on 32 large fallen trunks (diameter 60–130 cm) of pedunculate oak (*Quercus robur*) in various decay stages in the strictly protected zone of Białowieża National Park, Poland. The total number of species was 187, with 4–38 species per trunk. The mycobiota of individual trunks was unique, consisting of a variable set of several frequent species, a high number of infrequent to rare ones, and a considerable proportion of mycorrhizal fungi and species usually preferring conifer wood. Relations between trunk parameters, surrounding vegetation and fungus occurrences were analysed using multivariate statistical methods. The number of fungal species per trunk was significantly (negatively) correlated with the SW direction of trunk fall (which represents the highest heat load via forest canopy gap), and positively correlated with trunk size parameters, percentage of bark cover and contact with the soil. The species-richest trunks were those covered by bark, of large volume, in younger stages of decay, not exposed to heat from afternoon sun, and under a lower canopy cover. Trunks exposed to more intensive afternoon radiation had different and species-poorer macromycete communities than the shaded trunks characterised by e.g. *Hydropus floccipes*, *Mycena speirea* and *Pluteus podospileus*. The community composition also reflected trunk size characteristics, moss/bark cover of trunks and their contact with the soil. The presence of some dominants (*Ganoderma applanatum*, *Mycena inclinata*, *Kretzschmaria deusta*, *Xylobolus frustulatus*) had a significant effect on community composition. Some herbs requiring nutrient-rich soils occurred in the vicinity of more decayed trunks.



PS - poster session

**Microscopic fungi colonizing *Ursus spelaeus* bones and their susceptibility to commercially available antifungal preparations**

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Underground ecosystems are one of the most inhospitable places for microorganism development and functioning, mainly due to the lack of organic matter. Therefore, any organic matter applied to such area can stimulate their growth.

The main goal of this study was identification of fungi colonizing museum bone specimens of *Ursus spelaeus* in Niedźwiedzia cave (Poland). Moreover, in order to control the growth of fungi on bone exhibits, their susceptibility profile to commercially available fungicidal preparations was estimated.

The mycological analysis included both the surface and inner bone layers. The biological material was transferred to various agar plates (YPG, Sabouraud and BHI) using the dilution method. The plates were incubated at 10, 20 and 30±0.2°C for 7-28 days. Fungi were identified based on phenotypic tests and ITS region analysis. The antifungal activity of 3 preparations (Pufmax, Altax and Devor Mousse) was evaluated using serial dilution of the compound on agar plate and spot tests.

Phenotypic and molecular research showed that bones were colonized by 11 fungal species: *Absidia glauca*, *Aspergillus fumigatus*, *Chrysosporium merdarium*, *Fusarium cerealis*, *Mortierella alpina*, *Mucor aligarensis*, *M. plumbeus*, *Penicillium chrysogenum*, *P. expansum*, *Sarocladium strictum* and *Scopulariopsis candida*. All of the tested preparations were the most active against *C. merdarium*. In turn, *M. plumbeus*, *M. aligarensis*, *M. alpina* and *A. glauca* were the most resistant. The highest antifungal activity was shown by Pufmax (MIC and MFC of 0.16-0.63% and 1.25-2.50%, respectively). The lowest fungicidal effect was observed for Devor Mousse (MIC and MFC of 2.5-10% and 5-20%, respectively). Altax and Pufmax showed strong fungicidal activity at the concentration range of 1.25-5%.

PS - poster session

**Comparative metagenomic analysis of fungi inhabiting roots of *Deschampsia antarctica* and *Poa annua*, two grass species collected in Antarctica**

**Sebastian Pilsyk**, Urszula Perlińska-Lenart, Jakub Grzesiak, Anna Znój, Katarzyna Chwedorzewska, Marta Ajchler-Adamska Kamil Glizia Joanna S. Kruszczyńska  
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*Deschampsia antarctica* is a native Antarctic plant from the order Poales and Poaceae Family. The second species collected in the vicinity of Polish Antarctic Station “Arctowski”, *Poa annua* belongs to the same Family Poaceae, but unlike *Deschampsia*, *Poa* is one of the most ubiquitous plant species found around the World. It is already present at all continents and climatic zones.

The aim of this study was to analyze microbiome of the both grass species and conclude about possibilities of its involvement in adaptation of the plant to the harsh Antarctic climate conditions.

To analyze microbiome of fungi inhabiting roots of the two grass species DNA was isolated from rhizosphere, epirhizome and endorhizome. Fungal taxa were identified based on ITS1 rRNA region using primers ITS1FI2 and 5.8S. Sequencing was done using Illumina MiSeq Instrument and pair-end (2x250 bp) mode with V2Illumina kit. Singletons were removed (minimal OTU count = 10) and OTUs were picked with sequence identity criteria of 97%.

We have found significant differences in fungal microbiome of both grasses. Although Basidiomycota dominated in the rhizosphere and epirhizosphere of *Deschampsia*, while Ascomycota reached 76% of fungi inhabiting endorhizome. Opposite pattern of fungal representation was found in *Poa annua* where the epirhizosphere was colonized predominantly by Ascomycota, while inside of the roots Basidiomycota reached 82%.

These data will be supplemented with analysis of fungal microbiome of *Poa annua* collected from our climate zone.

PS - poster session

### **Aphyllorphoroid fungi of the Bialowieza Forest (Belarus)**

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Polish mycologist F. Błoński was the first who started to research the Bialowieza Forest (Błoński, 1887, 1889) but up to the middle of the XX century studying of the mycobiota was of irregular character. The extensive researches took place in 1950-70th years. They were done by the co-workers of the Scientific department of the National Park and Institute of Experimental Botany. For this period the Belarusian mycologists identified 159 species of the aphyllorphoroid fungi (Komarova, etc., 1968; Mikhalevich, 1971) and more than 300 species of the agaricoid basidiomycetes (Serzhanina, 1968).

In 2015-2018 the authors conducted new researches of the xylotrophic fungi of Bialowieza Forest (Belarusian part). All representative types of forest communities were surveyed. On the whole we have currently collected and deposited in MSK-F herbarium 79 genera and 140 species of the aphyllorphoroid fungi. It is less, than in the Polish part of Bialowieza Forest (Karasidski, Wochkowycki, 2015).

The most often found species were *Gloeophyllum odoratum* (Wulfen) Imazeki, *Fomes fomentarius* (L.) J.J. Kickx, *Fomitopsis pinicola* (Sw.) P. Karst., *Stereum hirsutum* (Willd.) Pers., *Trametes versicolor* (L.) Lloyd, *Ganoderma applanatum* (Pers.) Pat., *Xylodon paradoxus* (Schrad.) Chevall.

New localities of growth of the fungi from the Red List of Belarus *Cantharellus cinereus* (Pers.) Fr., *Fistulina hepatica* (Schaeff.) With., *Fomitopsis rosea* (Alb. & Schwein.) P. Karst., *Hericium coralloides* (Scop.) Pers., *Pycnoporus cinnabarinus* (Jacq.) P. Karst., *Sparassis crispa* (Wulfen) Fr. have been revealed.

As for the Belarusian part of Bialowieza Forest the species *Antrodia albida* (Fr.) Donk, *Hericium erinaceus* (Bull.) Pers., *Pycnoporellus alboluteus* (Ellis & Everh.) Kotl. & Pouzar were noted for the first time.



PS - poster session

**Macrofungi in birch bog forests on anthropogenically transformed raised bogs (NW Poland)**

**Małgorzata Stasińska**<sup>1</sup>, Zofia Sotek<sup>1</sup>, Ryszard Malinowski<sup>2</sup>, Renata Gamrat<sup>3</sup>, Małgorzata Gałczyńska<sup>4</sup>

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The raised bogs are highly susceptible not only to natural factors, such as unfavorable climatic changes but above all hydrological disturbances resulting from human activity. In Europe, almost all bog areas have been disturbed, whereas in Poland close to 80% of bog ecosystems have been subject to human interference. The study aim was to understand the mycological diversity of birch bog on raised bogs subject to anthropogenic changes. The research was carried out on 13 plots located on 6 anthropogenically modified bogs and on 2 plots on a non-disturbed peatland. Phytosociological and mycosociological relevés were taken and substrate analyses were carried out (pH, humidity, N-NH<sub>4</sub>, N-NO<sub>2</sub>, N-NO<sub>3</sub> and P-PO<sub>4</sub>). The research results indicate the existence of significant relationships between the occurrence of macrofungi and syntaxonomic group of plants and selected environmental factors, i.a. humidity and soil pH. A significant positive correlation was obtained between plant species of class Vaccinio-Picetea, and the total number of fungi species and bioecological fungi groups: mycorrhizal and saprotrophic (litter-inhabiting, growing on peat). Based on the obtained positive correlation between the total number of macromycetes on the given plot and certain bioecological groups of fungi, and the content of phosphorus and various nitrogen forms in the soil it can be expected that with their increase the macromycetes diversity will increase. The slow rate of the changes is demonstrated by the high diversity of fungi species, which was highest on the areas of former peat extraction pits, whereas lowest on bog margins, and particularly on the undisturbed bog.

PS - poster session

**Effect of deadwood on ectomycorrhizal colonisation of old - growth oak forests**

**Jacek Olchowik**<sup>1</sup>, Dorota Hilszczańska<sup>2</sup>, Roman Mariusz Bzdyk<sup>2</sup>, Marcin Studnicki<sup>3</sup>, Tadeusz Malewski<sup>4</sup>,  
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Although the importance of coarse woody debris (CWD) for species diversity is recognized, the effects of coarse woody debris decay class on species composition have received little attention. We examined how the species composition of ectomycorrhizal fungi (ECM) changes with CWD decay. We describe ectomycorrhizal root tips and the diversity of mycorrhizal fungal species at three English oak (*Quercus robur*L.) sites. DNA (ITS) barcoding revealed a total of 17 ECM fungal species. The highest degree of mycorrhizal colonization was found in CWD<sub>advanced</sub>(27.2%) and CWD<sub>early</sub>(27.1%). Based on exploration types, ectomycorrhizae were classified with respect to ecologically relevant soil features: The short distance type was significantly correlated with soil P<sub>2</sub>O<sub>5</sub>, while the contact type was correlated with soil C/N. The lowest mean content of soil C<sub>org</sub> was found in the CWD<sub>absent</sub> site. The difference in total soil N between sites decreased with increasing CWD decomposition, whereas total C/N increased correspondingly. In this study we confirmed that soil CWD stimulates ectomycorrhizal fungi representing contact or short-distance exploration types of mycelium.

PS - poster session

**Diverse endophytic fungi colonize indigenous grasses in the Hyrcanian Forest of Iran**

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Endophytes (endo=within, phyte=plant) are microorganisms that live within plants without causing apparent pathology. Endophytic fungi exist within most, if not all, wild plants, as they have done so since land plants first appeared 470 million years ago. In some cases, endophytic fungi are mutualistic partners, that is, they may provide benefits to their hosts under specific conditions of biotic or abiotic stress. In this study, we aimed to catalogue endophytes that colonize wild plants in one of the most unique and striking ecosystems in the world, the 40-million-year-old Hyrcanian Forest of Iran. Regions of the Hyrcanian forest were surveyed, and samples were collected from a range of indigenous grasses, especially poaceous species. Endophytic fungi were isolated from different parts of each plant, and pure cultures of each fungus were obtained using the hyphal-tip culture method. Total DNA of each isolate was extracted using a CTAB-based protocol, before internal transcribed spacer (ITS) regions of the ribosomal RNA gene were amplified and sequenced. Database searches indicated that the associated fungal endophytes were highly diverse within and between plants, with some displaying tissue-specificity. Fungal genera/species belonging to the families of Trichocomaceae, Nectriaceae, Pleosporaceae and Sordariaceae were identified. While some fungal isolates represented novel species, some others could not be classified to the level of genus or even family, indicating that there is considerable unexplored diversity within the endophytes colonizing the plants in this region. The roles that these endophytes may play in the biology and ecology of their hosts promises to be a fertile area of future research, with potential implications for agricultural systems.



PS - poster session

**A new ecosystem and its fungal community: exploratory study of a pristine climacic forest patch in Southern Portugal**

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Recently a novel Mediterranean forest association, represented by a few remnant patches in southern Portugal, has been proposed. It is a climacic forest that can occur along streams, dominated by *Quercus faginea* subsp. *broteroi* (Cout.) A.Camus (= *Quercus broteroi* (Cout.) Rivas Mart.), but in contrast to other oceanic associations of the same species, it occupies drier areas in transitional edaphoclimatic envelopes. This pristine vegetation type is vulnerable, due to its highly fragmented, limited expanse. The results of Fall-Winter macrofungal forays in a well preserved patch, from 2014 to 2018, are presented here, documenting a species-rich assembly radically different from the ones associated with other oak species in the same region. On the other hand, they bear some resemblance with the results from other forest types dominated by *Q. faginea* s.l. in the Iberian Peninsula. The temporal dynamics of sporocarp production, in relation to climate variables, is also analysed. Our study highlights the critical importance of protecting this vulnerable ecosystem, regarding local land use decisions as well as global climate change.

PS - poster session

**Fungal Diversity in Sacred Groves vs. Managed Forests of Epirus, NW Greece**

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Sacred groves in Epirus, NW Greece, were investigated for fungal diversity compared with nearby managed forest, similar in vegetation. Sacred groves are protected by religious respect, remain undisturbed and are comprised of old trees. Plots in both sacred groves and managed forest (control) were set out in 8 sites. Sampling was conducted during the fall of 2013, 2014 and 2015 following a sampling protocol. The 180 species of macrofungi recorded in all plots were classified into 4 ecotrophic groups: mycorrhizal, saprotrophic, xylotrophic and parasitic. When species and populations were considered as a whole, sacred groves appeared to shelter a higher number of species and larger populations. When the ecotrophic groups were compared individually, the number of species and the populations of mycorrhizal fungi were found to be higher in the control plots of four sites than in their corresponding sacred plots. In three other sites, there were no significant differences and only one sacred plot was significantly higher. On the contrary, saprotrophic fungi appeared to be higher in number of species and density in sacred plots of five sites while in one site they were the same. In the two remaining sites the control figures were higher. The group of xylotrophic fungi was significantly richer in the sacred plots of six sites, while in the remaining two sites, the control plots were richer. In the group of parasitic fungi, evaluation of incidence and abundance was not possible because the most common parasitic fungus, (oak mildew - *Erisyphe alphidoides*), was not countable. In conclusion, sacred sites hold more saprotrophic and xylotrophic fungi because of the abundance of old trees, while mycorrhizal fungi thrive in managed forest with younger, fast growing trees.

PS - poster session

### **The soil microfungi of the Colombian ecosystems**

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Although Colombia is one of the first tropical countries where an effort was made to gather a mycological flora, contributions to the taxonomy, diversity and ecology of soil microfungi are still scarce. Study on microfungi has increased only in the last decades and the research focuses especially to functional groups, metabolites and role in the agroecosystem.

In this study the diversity of soil microfungi is analysed comparing data from the literature in relation to the natural regions of the country: Andean, Amazonian, Caribbean, Orinoquía, Pacific and Insular. The analysis shows that most of the data refer to the Andean, while the Pacific region is the least studied. No data are reported for the Colombian Insular region. The high diversity reported for the Andean may be due to its extension, eco-geographical diversity and to the fact that it is the most populated region, and accessible to research activity. No more than 170 species of soil microfungi for Colombia have been recorded, corresponding to 58 genera and 3 phyla (Ascomycota, Zygomycota and Glomeromycota). Referring to the soil fungi of the country, it has been possible to determine that arbuscular mycorrhizae represent the most widely studied group. The data from this review allow to affirm that the most recorded genus in Colombian soils is *Glomus* with 38 species, followed by *Acaulospora* with 13 species, *Penicillium* with 12 species, *Aspergillus* and *Fusarium* each with 10 species and *Mortierella* and *Trichoderma* each with 7 species. The other genera appear in lesser proportion and with less diversity of species in the soil. The information collected on soil fungi from the literature are analysed with recent data recorded from the Andean region by means of traditional method coupled with metagenomics analysis.

## HYPOGEOUS MYCORRHIZAL FUNGI

OS - oral session

**Competitive dynamics in *Tuber* genus: what do we know?**

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Truffle in the genus *Tuber* are mutualistic ascomycetes that form ectomycorrhizas (ECM) with a wide range of plants. *Tuber* spp. are native of the northern hemisphere but human activities contributed to introduce them, accidentally or by cultivation, in the southern hemisphere. The genus exhibits high continental endemism and provincialism and most of the species preferentially grow in calcareous alkaline soils together with many other pezizalean ECM fungi. However, some species also grows in acidic soils where *Cenococcum geophilum*, Russulaceae and Cortinariaceae are more frequent. The ecological diversity in the *Tuber* genus makes possible to find its members in a broad range of forest biomes.

In the wild, *Tuber* ECMs are patchily distributed and rarely dominate the ECM fungal community. On the contrary, they dominate the community of man-made truffle plantations for a longer or shorter time depending on the fitness of the introduced species to the ecological conditions of the site.

Usually, mycelia of different *Tuber* species can co-occur in the same bulk soil in habitats suited for truffles but the competition for a space on the host roots is often severe between them. The competitive exclusion between strains of the same species seems also to be consistent in *Tuber*. Field studies conducted on different *Tuber* species showed that single or few genotypes dominate on the roots of distinct plants. Probably, the presence of severe (and still unknown) vegetative incompatibility systems prevents the colonization by other genotypes, in particular those of opposite mating type.

The understanding of competitive dynamics affecting the permanence of *Tuber* members within an ECM community is one of the challenges for improving truffle cultivation strategies.

OS - oral session

**The Algerian desert truffles: taxonomy, distribution and mycorrhizal ecology**

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Morphological and phylogenetic analyses of large ribosomal subunit (28S rDNA) and internal transcribed spacer (ITS rDNA) of desert truffles samples collected from several bioclimatic zones in Algeria revealed the presence of five distinct *Terfezia* species: *T. arenaria*, *T. boudieri*, *T. claveryi*, *T. eliocrocae* and *T. crassiverrucosa*. The molecular investigations within the *Picoa* genus highlighted several genetic lineages which appear to be separated more efficiently by geographical and ecological data, rather than morphological characters. *Tirmania* is the third genus of desert truffles present in Algeria. Desert truffles species are exceptional in terms of mycorrhizae structural plasticity, as they are able to produce sheeting ectomycorrhizae, endomycorrhizae or even ectendomycorrhizae with Cistaceae hosts and Aleppo pine.

OS - oral session

**Hypogeous fungi in the Republic of North Macedonia – what we know and what can we expect**

**Slavica Tofilovska**<sup>1</sup>, Katerina Rusevska<sup>1</sup>, Mitko Karadelev<sup>1</sup>, Tine Grebenc<sup>2</sup>

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Hypogeous (or sequestrate) fungi are important ecosystem element, many genera are mycorrhizal, thus playing significant role in the forest ecosystem functioning. The diversity of hypogeous fungi still remains understudied in the Balkan Peninsula, in particularly outside traditional truffle hunting areas. The Republic of North Macedonia is located in the central part of the Balkan Peninsula, on its territory the high geological diversity and effects of different climate types shaped a rich biodiversity and suitable ecosystems for thriving of hypogeous fungi. Until recently, the occurrence data for hypogeous fungi in RNM was scarce, with overall only 18 species recorded (Chavdarova et al., 2011, Marjanovic et al., 2010). However, the intense collaboration with truffle hunters and specialists resulted in registering higher number of hypogeous fungi, including 14 taxa from the genus *Tuber* and additional species from the genera *Rhizopogon*, *Hymenogaster* and *Hysterangium* (Karadelev et al., 2018, 2019). In this work we are presenting 10 hypogeous taxa for the first time registered in RNM: *Gautieria morchelliformis*, *Hymenogaster citrinus*, *H. vulgaris*, *H. populetorum*, *Melanogaster ambiguus*, *M. macrosporus*, *M. umbrinileba*, *M. variegatus*, *Pachyphloeus conglomeratus* and *Stephensia bombycina*. We prepared distribution maps for all recorded taxa, depicted on the map for regional biogeographic division (Melovski et al., 2013) as overview for the richness of different regions in RNM. Ecology, habitat preferences and potential or confirmed mycorrhizal partners will also be presented. Comparing the record abundance with neighboring areas we believe the number of recorded taxa is still low and a need for detailed exploration of specific ecosystems will be discussed.

OS - oral session

**Shedding light on the *Tuber mesentericum* complex**

**Marco Leonardi<sup>1</sup>**, Mirco Iotti<sup>1</sup>, Gian Luigi Rana<sup>2</sup>, Aurelia Paz-Conde<sup>3</sup>, Daniele Salvi<sup>1</sup>, Giovanni Pacioni<sup>1</sup>

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*Tuber mesentericum* Vittad. is an edible European black truffle of some commercial interest, apparently easy to recognize due to its warty peridium, excavate base, reticulate spores and frequent phenolic-like odor. The species is widely distributed across Europe and is very abundant in central-southern Italy. This species lives mainly on calcareous soils and forms ectomycorrhizas with different host trees. The taxonomy of *T. mesentericum* was early affected by several criticisms. A few taxa similar to *T. mesentericum* were described over the time (e.g. *T. bituminatum* Berk. & Broome, *T. cibarium* Corda, *T. culinare* Zobel) but different authors do not consider them as valid. Indeed, the ascomata of these fungi display homogeneous morphological features, often shared with the closely related *Tuber aestivum* Vittad. Phylogenetic studies based on *T. mesentericum* ITS sequences showed the occurrence of a species complex with at least three putative cryptic species. Currently it is not yet clear which of these phylotypes correspond to the “true” *T. mesentericum*, also because its holotype is missing. In order to clarify the phylogeny of this truffle, we analyzed a specimen voucher of Vittadini and other specimens designed as holotype of *Tuber bituminatum* by Berk. & Broome, both from the herbarium of C. E. Broome preserved at KEW. The poor quality, the old age (> 150 years) and the low amount of available tissue of these specimens forced us to design a set of specific primer pairs targeting the most informative part of ITS region. In addition, we attempted to clarify the relationships between the cryptic species belonging to *T. mesentericum* complex by a multi-locus genomic analysis and we designated a neotype of this taxon based on ascoma morphology and ITS rDNA sequence data of an authentic specimen from Vittadini.



OS - oral session

**Insects inhabiting fruiting bodies of Burgundy truffle (*Tuber aestivum* Vittad.)**

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The aim of this research was to provide a survey of the insects that may be associated with the fruiting bodies of Burgundy truffle (*Tuber aestivum* Vittad.) in Poland. The study was carried out in 2016-2017 at four natural truffle sites located in the southern Poland. The fruiting bodies were searched with the help of trained truffle dog, Lagotto romagnolo breed, or randomly collected. Adult insects were sampled in the sites, directly from the fruiting bodies, with help of an exhaustor. The fruiting bodies potentially inhabited by insect larvae were collected, transported to the laboratory and placed in special containers. The insects' rearing was carried out in biological chamber with constant temperature and humidity parameters. Both, adult insects collected in the sites and those reared in the laboratory were identified using morphological methods.

In time of the research 183 fruiting bodies were found and investigated. In total, 330 insect specimens belonging to 21 species were identified. For the first time in Poland presence of *Suillia gigantea* (Meigen, 1830) (Diptera) was confirmed, based on two specimens of the species, which were bred from truffle fruiting bodies. The assemblage of insects inhabiting truffles has been identified, with two dominants: flies belonging to the Syrphidae family and truffle beetles of Leiodidae. Some of the species can cause significant losses in truffle orchard. Others can be considered as indicators of truffle occurrence in the forest environment. The pioneering research conducted in Poland has contributed to a significant increase in knowledge about insects inhabiting truffles, their occurrence and biology.

OS - oral session

**Small mammals as dispersal agents of hypogeous fungi in the Pieniny Mountains,  
Western Carpathians**

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The role of mammals as dispersal vectors of hypogeous fungi spores is understudied in Europe. We investigated the presence of hypogeous fungi in the diet of small mammals in the Pieniny Mts, Western Carpathians, in July and September 2016 and 2017. We hypothesized that: (1) high diversity of fungal sporocarps recorded in the Pieniny Mts is reflected in the diet of animals, (2) spores are transported outside forests by animals but both, the diversity and quantity of fungal spores delivered are lower than those observed within forests, (3) mycophagous mammals are not selective for consumed fungi. The diversity of consumed fungi in the Pieniny Mts was found to be very high: spores of over 30 taxa from 16 genera were present in nearly 70% of faecal samples of nine animal species analysed. Both, spore diversity in samples and the frequency of spore-containing faeces differed; however, between animals, years and between seasons. The highest mean number of fungal taxa per sample and the highest percentage of positive samples was recorded in the case of the bank vole, *Myodes glareolus*, and the yellow-necked mouse, *Apodemus flavicollis*. We showed that fungal spores were transported to meadows most effectively by the yellow-necked mouse. However, both, the diversity of taxa and frequency of spore-containing faeces were lower for samples collected in meadows than those collected in forests. For the first time our investigation proved that the herbivorous rodent, the common vole, *Microtus arvalis*, shows opportunistic mycophagy. Our field-based results suggested differential consumption of hypogeous fungal species by specific rodents. A laboratory experiment performed with the use of the bank voles proved some degree of specialization in the choice of this mycophagists for fungi.

PS - poster session

**Community composition of root-associated fungi in a productive *Tuber aestivum* Vittad. orchard**

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In terrestrial ecosystems plant roots are colonized by mycorrhizal, endophytic, saprobic and pathogenic fungi. Focused on the root systems of common hazel (*Corylus avellana* L.) and pedunculate oak (*Quercus robur* L.) in a first productive truffle orchard in Poland, we used next generation sequencing (NGS, Illumina) to explore how phylogenetically diverse fungi constitute an ecological community of two different host-plants. In total, 134 operational taxonomic units (OTUs) of fungi were found from 20 collective terminal-root samples from two plant species occurring in the orchard. Among the all fungal OTUs found from the samples, 133 (99,8%) were identified at phylum, 127 (94,8%) at order and 106 (79,1%) were identified at genus level. At phylum level, 72 (54,1%) fungal OTUs belong to ascomycetes, 54 (40,6%) to basidiomycetes, four to glomeromycetes (3,0%), two were mortirellamycetes (1,5%) and one was rozellomycetes (0.7%). The root-associated fungal community was dominated by ectomycorrhizal fungi of Pezizales, Thelephorales and Agaricales. Diverse ectomycorrhizal clades were observed, viz. *Tuber*, *Inocybe*, *Russula*, *Cortinarius*, *Tomentella*, *Boletus* and *Cenococcum*. Among the fungi classified as endophytes, these from the order Helotiales were most common. .

Overall, this study revealed that root-associated fungal communities present in truffle orchard were dominated by ectomycorrhizal fungi and diverse root endophytes as well. The potential ecological interactions between the two host-species may be important to understand the complex assembly processes of belowground fungal communities.

PS - poster session

**The neglected hypogeous ectomycorrhizal fungi of Artikutza (Northern Navarre, Spain)**

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The beneficial roles that hypogeous ectomycorrhizal fungi perform in forest ecosystems have been occasionally assessed (Claridge, 2002; Izzo et al. 2005). However, these fungi are commonly neglected in fungal studies, particularly in temperate forests, as opposed to epigeous ones. Among other reasons, it is worth highlighting the difficulty of inventorying them, although molecular methods have partially alleviated this problem. We present here a hypogeous ectomycorrhizal community analysed, at the level of root tips and sporocaps, in a study carried out in a beech forest in Northern Spain (Artikutza, Navarre). The species richness on the roots was characterized by a combination of morphotyping under a dissecting scope and subsequent DNA sequencing the ITS region. We used specially trained dogs to sample the hypogeous fungal community. A total of 26 taxa have been identified, including two newly described species (currently under study). But the number of taxa found within each method was lower and similar (16), with only 50% of them being shared in both analyses. Despite being the most typical species of beech, they are rarely recorded in the Basque Country, and some are new records to the area, such as *Luteoamylascus* sp. With the data analysed so far, it should be noted that the number of hypogeous ectomycorrhizal species is significantly higher in trees located in old iron mines compared to trees in undisturbed areas. This result may be in accordance with data from other studies that point to a more noticeable presence of hypogeous in more stressed areas.

PS - poster session

**Biogeography and ecology of *Tuber aestivum* in Croatia**

Magdalena Jambrek Fuellhart<sup>1</sup>, Ana Pošta<sup>1</sup>, Željko Zgrablić<sup>2</sup>, Dino Buršić<sup>2</sup>, Ivana Kušan<sup>1</sup>, Neven Matočec<sup>1</sup>,  
Zdenko Tkalčec<sup>1</sup>, **Armin Mešić<sup>1</sup>**

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The Republic of Croatia is a small European country which spans to four different biogeographic regions and a large number of bio-climatic belts. Due to the significant differences between those, many forest habitats are developed within a small area. The initial results of the first ongoing comprehensive study on hypogeous mycorrhizal fungi in the forest ecosystems of Croatia, regarding a widespread commercial truffle species - *Tuber aestivum*, are presented here. The importance of commercial truffles in the Istrian peninsula (including *T. aestivum*) is summarized by Zgrablić et al. (2014a, 2014b). There are few earlier published records on hypogeous fungi in Croatia, starting with the pioneer work of Sella (1932). Although *T. aestivum* has been commercially exploited here for many decades, the available data remained scarce, often unreliable and thus scientifically unprocessed. The main aim of this research was to examine the biogeographic and ecological features as well as the distribution extent of *T. aestivum* within a frame of diversity of natural and seminatural habitat types in Croatia. The analysis of ca. 100 collections deposited in the Croatian National Fungarium (CNF) showed that *T. aestivum* is most common in native forests with *Quercus ilex*, *Q. pubescens*, *Q. cerris*, *Carpinus orientalis*, *Ostrya carpinifolia* in the Mediterranean region, and *Fagus sylvatica*, *Carpinus betulus*, *Quercus petraea*, *Corylus avellana* in the Continental region, developing mature fruitbodies mainly from May to December. The results of this research could provide reliable data about natural productivity of the species on a national scale for the first time. The nomenclature of *T. aestivum* is discussed.

PS - poster session

**Diversity of fungi inhabiting resin tapping wounds of Scots pine in Latvia: preliminary results**

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Scots pine (*Pinus sylvestris* L.) is one of the most economically important conifer species in Baltic region. Pines are among the most commercially important tree species valued for their timber. Since 19th century approximately 20 000 ha of Scots pine stands in Latvia have been used in resin tapping. Although resin exudation is the part of defence mechanism against fungal and insect attack, huge wounds are subjected to infection by stem decay causing fungi.

The aim of this study was to evaluate the decay incidence and associated fungi in *Pinus sylvestris*. More than 30 years after resin tapping. In three stands trees were evaluated for resin tapping wounds and 30 trees in each stand were selected. In total 127 resin tapping wounds of 90 pines were measured. One wood sample was collected 1 cm bellow injury for evaluation of presence of decay and subsequent fungal isolation. Isolates were categorized as morphotypes. One to two representatives of each fungal morphotype were subjected to molecular identification. In total 47 fungal taxa was obtained, from those six species of basidiomycetes were isolated. Majority of isolated fungi were saprotrophs or endophytes; and only few were pathogens.

PS - poster session

**Diversity and distribution of genus *Tuber* in the Republic of North Macedonia**

**Slavica Tofilovska<sup>1</sup>**, Katerina Rusevska<sup>1</sup>, Mitko Karadelev<sup>1</sup>, Tine Grebenc<sup>2</sup>

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Studies on hypogeous fungi in the Republic of North Macedonia (RNM) was intensified in the last decade and until recently only 5 taxa of genus *Tuber* were noted (Marjanović et al., 2010). Due to the commercial value of genus *Tuber*, more records are being generated and all together 17 taxa were registered for this genus in RNM (Karadelev et al., 2019). Review of available literature and databases for RNM show that most of the common *Tuber* taxa were collected: *Tuber aestivum*, *T. borchii*, *T. brumale*, *T. excavatum*, *T. mesentericum*, *T. puberulum*, *T. rufum* f. *ferrugineum* and *T. rufum* f. *lucidum*; all with wide distribution and abundant recorded sites. Additionally, taxa with limited distribution in RNM were also retrieved: *Tuber brumale* f. *moschatum*, *T. foetidum*, *T. fulgens* and *T. rufum* f. *nitidum*. Finally, *Tuber bellonae*, *T. dryophilum*, *T. macrosporum* and *T. magnatum* only had one record or one site where they were recorded consecutively. Among cultivated truffle species, only *Tuber melanosporum* is successfully cultivated and is not found in a natural habitat. In RNM the most frequently marketed truffles are *Tuber aestivum*, *T. borchii* and *T. brumale*, while *T. magnatum* remains underexploited or rare, mainly due to the degradation of its natural habitats in the past. The recent descriptions of new species from the Balkan Peninsula, e.g. *T. petrophilum* (Milenkovic et al. 2015) and *T. pulchrosporum* (Polemis et al. 2019) suggests that handful of hypogeous diversity, including the genus *Tuber*, in the Balkan Peninsula still remains unexplored.

PS - poster session

### **Hypogeous fungal diversity in Serbia**

**Boris Ivančević**<sup>1</sup>, Miroljub Milenković<sup>2</sup>

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The oldest data on hypogeous fungi from the territory of Serbia date back to the year of 1777, but very little was known about them to this day. Small number of published articles and papers primarily addressed the true truffles (*Tuber* spp.), and offered almost no data about other groups, which presented a large unknown component of Serbia's fungal diversity. Investigation of hypogeous fungi with the help of trained dogs led to the formation of a collection of nearly of thousand specimens. As many as 70 taxa were registered, indicating a far greater richness than previously recognised. The data on distribution of these species were analysed with the aim of assessing correlations with the types of habitat and other ecological factors. The study gives ranges of variation of ecological parameters, degrees of habitat dependence, values of total and assessments of potential diversity, as well as dependence on bioclimatic variables for all species. It provides assessments as to the factors of threat, and identifies potentially the most valuable areas for conservation.



PS - poster session

**Morphological and DNA characterization of *Tuber panniferum* ectomycorrhizae**

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*Department of Agricultural, Food and Environmental Sciences, Perugia, Italy*

*Tuber panniferum* Tul. & C. Tul. is a rare truffle, apparently linked to the Mediterranean climate. Ectomycorrhiza (ECM) of *T. panniferum* can be easily confused with those of most known black truffles *T. melanosporum* Vittad. and *T. aestivum* Vittad. that are marketed and cultivated all over the world. Therefore, to avoid misidentification of *T. panniferum* with these two species, both morphological and molecular analysis for ECMs are recommended. In this study 24 seedlings of *Quercus pubescens* Willd. were inoculated blending approximately 38g of a fresh *T. panniferum* ascocarp and 8 seedlings were included as control. Seedlings were placed in a growth chamber and grown for one year. The level of ectomycorrhizal colonization was assessed after 8, 9 and 12 months. At all the three check-points at least one plant presented *Tuber*-like ECMs belonging to the same morphotype. The ITS region of three ECMs was sequenced and recognized as belonging to the *T. panniferum* species. The ECM of *Tuber panniferum* was described in this study. The ECM is club-shaped, brown at complete formation and densely covered by the emanating hyphae. Not ramified cystidia are present in the outer mantle surface, which is formed of angular cells organized in a pseudoparenchymatous tissue. These evidences showed a high similarity among the ECM of *T. panniferum* and *T. aestivum*. Moreover, our description has some differences with that proposed by Pirazzi in 1998, who didn't provide a molecular characterization of the species.

This study renews the description of *T. panniferum* ECM, with the support of molecular data. Our results shed light on the similarity among *T. panniferum* and *T. aestivum* ECM, highlighting the importance of an accurate identification to avoid any misclassification.



PS - poster session

**Microbiome of fruiting bodies of Burgundy truffle (*Tuber aestivum* Vittad.)**

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*Tuber* species form ectomycorrhizae with trees and shrubs. Some of them produce edible fruiting bodies with a unique flavor and texture. Two species, *Tuber magnatum*, the white truffle, and *Tuber melanosporum*, the black truffle, are the most valued but their natural habitats are limited to Italy, France, Spain, Croatia, Slovenia and Hungary. Comparing with the two species, *T. aestivum* is much wider distributed in Europe. In this study we have used *T. aestivum* collected in Poland.

*Tuber* species hosting diverse microorganisms inside their fruiting bodies. In this study, we identified bacteria and fungi inhabiting the gleba of *T. aestivum* using culture dependent and metagenomic analysis.

Analysis of the V3-V4 region of 16S rDNA identified nine phyla of bacteria present in the gleba of *T. aestivum* ascomata, mostly Proteobacteria from the family Bradyrhizobiaceae.

The ITS1 region of fungal rDNA represented, apart from *T. aestivum* itself, six fungal species belonging to three phyla, Ascomycota, Basidiomycota and Mucoromycota.

To complement the metagenomic analysis, cultivable fungi and bacteria were obtained from the gleba of the same *T. aestivum* fruiting bodies. The identified fungi mostly belong to the phylum Basidiomycota and same to Ascomycota. Notably, one fruiting body was found to be almost devoid of alien fungi by both approaches, and for another fruiting body both analyses showed that it carried the most diverse bacterial community.

In general, however, the results of the culture –dependent analysis and of the metagenomic approach were completely different and complemented each other, indicating that both methods have their own merit and should be used in combination to afford full information on complex microbiota.

PS - poster session

### **Evaluational approach to the Red List of hypogeous fungi in Poland**

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The IUCN Red Lists edited at regular intervals contain surveys of species belonging to different groups of threatened organisms on a global scale. The Global Fungi Red List Initiative propose to assess and document at least 1500 fungi by 2021. A special Practical Workshop assessing the global conservation status of fungi took place e.g. during last meeting of European Council for Conservation of Fungi in Macedonia (Dahlberg 2017). Some complications of using IUCN Red List criteria are discussed when assessing hypogeous fungi. They present a special ecological group of macromycetes forming their fruit bodies underground and remain in soil until they break down naturally. They occur in a large variety of habitats from virgin forests to sites in the city parks. The real status of threat has not been recognized, because only a few, if any, people in country specialise in this group of fungi and random collection cannot be a basis for the evaluation of threat.

As we can see from the analysis of European Red Lists the strategy of inclusion of hypogeous fungi varies between to extrema: first of them is to include all collected hypogeous fungi on the basis of their rarity. The second extremum remains to not include them at all because of deficiency of our knowledge and lack of data. The fundamental question is which fungi known as rare species are really rare and which are only under-recorded?

Some examples of assessing conservation status of hypogeous fungi from genera: *Elaphomyces*, *Tuber*, *Chamonixia*, *Octaviania* and *Melanogaster* are the matter of presentation.

Research financed by the Polish Science Centre (grant no. N N305 3045 40).

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PS - poster session

**TRUE – the revived TAUESG. Truffle Research Union of Europe for a new European truffle perspective**

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The network TAUESG, the *Tuber aestivum/uncinatum* European scientist group, was formed in 2009 during its first meeting in Vienna. The idea was to gather scientists with an interest in *Tuber aestivum/uncinatum*, which despite its value, its broad ecology and the fact that it was known to occur naturally in most European countries was underestimated in its potential and likewise under-researched compared to other more commercial species. The main goal of the group was thus to fortify science and practice on this species through collaboration between group members.

Gérard Chevalier was the first president and the group was constructed of the representatives of 17 European countries, at that time. With time, more European countries joined, but also non-European representatives from USA, Israel, New Zealand, Morocco and China participated in meetings. At the conference in Nancy (France) in 2011, 23 European and four non-European countries were represented. The open character of the meetings allowed not only scientists, but also seedling producers, growers and other people devoted to truffles to attend. Over the past ten years, the *T. aestivum/uncinatum* connected science and practice has spread all over the continent, and the goal of TAUESG has been achieved. During the last years, the enrolment in the group started to decline. This decline was broken by the seventh meeting of the group held in November 2018 in Visby, Sweden, where high impact scientific progress was presented and brought back the good spirit and the enthusiasm of the participants. It was especially emphasized that there is a need for enhanced investigation and regulation of non-traditional truffle regions and markets in Europe - Central, Eastern and Northern Europe and the Balkan Peninsula. The group topic has become attractive for research on ectomycorrhiza in general, which brought new perspectives for collaboration.

A transformation of the scope of the group was proposed and the outcomes of the discussion were that the group should also include research on other *Tuber* spp. A new acronym and leadership model were proposed with the name Truffle Research Union of Europe (TRUE), associating not only to the union of European scientists working on truffles, but also of the union of science and practice towards better collaboration, communication and application of scientific results. A new president was elected, Dr Žaklina Marjanović (University of Belgrade, Serbia) and the vice president, Dr Christina Wedén (Uppsala University, Sweden). The organizational structuring of the new group is in progress. The first official meeting of TRUE is planned to be organized in Serbia in 2020.

## FUNGAL CONSERVATION

OS - oral session

### **Advancing Fungal Conservation**

**Gregory Mueller**

*Chicago Botanic Garden, Glencoe, USA*

Fungal conservation is progressing rapidly! 100 fungi are included on the most recent IUCN Global Red List with another 150 or more assessments scheduled for publication later this year. National and regional red lists are being created or updated throughout the world. Projects implementing conservation actions to mitigate the loss of species are being initiated. And, fungi are increasingly being considered in policy and management plans. These are all very positive steps, but more is needed. While having several hundred species on the IUCN Global Red List is a major increase over the 3 species that were listed 5 years ago, it is far too few. Less than 0.025% of described species have been globally assessed. Listing species is a critical first step, but more conservation action plans are needed to mitigate the loss of threatened species. These conservation action plans should be based on the best data available. May et al. 2018 on conservation mycology provides a list of research questions relevant to fungal conservation. Lastly, but importantly, fungi are not yet part of the mainstream of conservation. Too often fungi are left out of conservation discussions, planning, and funding even though there is a growing body of data on fungal ecology and successful management practices to give useful advice for including fungi in conservation initiatives. Communicating the importance of fungi, documenting that fungi can and need to be included in conservation efforts, and volunteering time and knowledge to requests for engagement and information are critical ways that members of the mycological community can advance fungal conservation. This is an exciting time for fungal conservation, with continued effort and more people engaged much more progress can be made.



OS - oral session

**Fungal conservation in practice – 40 years' experiences from the Nordic countries**

**Anders Dahlberg**

*Swedish University of Agricultural Sciences, Uppsala, Sweden*

Over the last 40 years, fungi have risen from insignificance to approaching almost sharing equal prominence with animals and plants in Finnish, Norwegian and Swedish nature conservation. This is largely due to the pivotal role played by the national Red Lists as a widely accepted official document and knowledge source which is used for setting priorities and driving conservation actions. They are comprehensive, covering animals, macrofungi and plants, and it reports on status, trends and how rarity and decline can be mitigated.

The national Red List in Norway and Sweden is revised every five year; in Finland every ten year. In each country, on average 750 macrofungi are listed as red-listed by employed mycologists and their associated specialist groups – comprising 15% of all the nationally species red-listed. In Sweden and Norway, Species Information Centres produce these Red Lists and provide hubs for authorities, landowners, scientists, amateurs and other stakeholders to analyse, synthesize and spread knowledge of species and habitats. Their overall aim is to facilitate nature conservation.

The fungal presence in the Red Lists have clearly raised the interest and awareness of fungal diversity and fungal conservation. Fungi are accounted for when areas for protection are selected and in conservation initiatives and measures, e.g. in guidelines for environmental consideration such as setting aside Woodland Key Habitats. To an increasing extent, fungal conservation issues are researched and results implemented. The professional and private individuals contribute significantly for nature conservation by reporting fungal observations to national species websites (presently ca 2 million finds).

OS - oral session

**Deployment of NextGen sequencing in fungal conservation**

**Gareth Griffith**

*Aberystwyth University, Aberystwyth, UK*

The profile of fungal conservation in Europe has steadily increased over recent decades and by now several sites have legal protection based on the distinctive/rare macrofungal populations which inhabit them. For certain habitats in the UK, surveys of fungal diversity can be a requirement for EIA (Environmental Impact Assessment) in planning applications or proposals for agricultural intensification. When such surveys are required, there can often be costly delays (until fruiting season), and the ephemeral nature of macrofungal fruiting can require several site visits. We have developed a method for soil eDNA analysis to assess populations of grassland fungi. This has been used to identify grasslands at a range of UK grassland sites to resolve planning application and in one case use of eDNA was instrumental in conferring legal protection to one site (Leasowes in Birmingham) -a worldwide first in the use of eDNA to provide legal protection in nature conservation.

We have also designed low-cost airsamplers to trap airborne propagules for DNA metabarcoding. We have deployed this in the detection of the rare fungus *Cryptomyces maximus* (on IUCN top 100 threatened species) and also for biodiversity assessments in a range of habitats.





OS - oral session

**Medicinal properties of macromycetes in Shikahogh State Reserve of Armenia**

**Lusine Margaryan<sup>1</sup>**, Vahagn Gevorgyan<sup>2</sup>, Yeva Hovhannisyan<sup>1</sup>, Siranush Nanagulyan<sup>1</sup>

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Biological diversity is one of the powerful reservoirs of resources. The main tasks of a modern science is the study and preservation of biodiversity and its conservation in separate regions including special protected nature areas, which is one of the main key for humanity sustainable development. The goal of this work is a comprehensive study of macrofungi species composition in Shikahogh. The reserve (12.137 ha) is located in the southeastern part of Armenia. As an experimental material was chosen macroscopic fungi collected from the territory of the Shikahogh Reserve as well as the samples from the herbarium (ERCB) of the Department of Botany and Mycology of Yerevan State University. The studies were carried out during the vegetation period using the route-expedition method. Within the studied area 436 species of macrofungi of Ascomycota and Basidiomycota were revealed. In our studies we also registered 99 species of macromycetes with medicinal properties. They belong to classes of Agaricomycetes, Sordariomycetes and Tremellomycetes and 9 orders. The main part of the fungi with medicinal properties refers to the order of Agaricales (52 species). Most of the species found on the territory of the Reserve have antibacterial, antiviral, antifungal properties which are associated with such chemicals as terpenoids, purines, phenol derivatives. According to our data, the local population engaged in animal breeding uses *Lycoperdon perlatum* for the treatment of purulent diseases of rabbit ears. Thus, this study indicates that the collected data can be used for an update of the data passports of protected areas in Republic of Armenia and it is giving potential opportunities for the usage of macromycetes in medicine and the national household.



OS - oral session

**What do polish students know about fungi? A study on mycological education in primary school and nature education centers**

**Igor Siedlecki**, Michał Kochanowski, Alicja Okraśńska, Julia Pawłowska, Marta Wrzosek  
*Department of Molecular Phylogenetics and Evolution, Biological and Chemical Research Centre, Faculty of Biology, University of Warsaw, Warsaw, Poland*

Although Polish people are commonly perceived as fungi enthusiasts, common knowledge about fungi in Poland is limited mostly to edible mushrooms. Society's insufficient mycological knowledge may be a result of the limited amount of mycological content in school curriculum and in informal education. To check this hypothesis we decided to study: what primary school students learn about mycology, if the school curriculum covers up-to-date knowledge about fungi and how much information about each kingdom is presented in textbooks. In order to do that we decided to analyze the national curriculum and textbooks for biology classes in primary school, as well as the curriculum of 10 nature education centers. Additionally, together with mycologists from Polish universities, we created a list of 10 mycological topics that students should learn in primary school. The results suggest that although the national curriculum covers all mycological topics that appeared on the list, mycological content is just a small percentage (4%) of all biology taught in primary school. Both the national curriculum and the textbooks are focused on animals and plants. In the textbooks which focus on mycology and botany, botanical issues appear on average in 90% of subsections while fungi appear in only 30%. Moreover, fungi are used more rarely than animals or plants as examples to explain broader biological topics (e.g. Evolution). Finally, nature education centers also focus mostly on animals and plants. On average, only 17% of their classes include mycological content. The results of our studies show that while the school curriculum provides an opportunity to teach broadly about mycology, fungi are still marginalized in the biology textbooks and in the curriculum of informal education centers.

OS - oral session

**Celebrating the Fungi to fight fungal blindness and boost conservation**

**Susana C. Gonçalves**<sup>1</sup>, Miguel Ferreira<sup>1</sup>, Joana Cabral-Oliveira<sup>2</sup>, António C. Gouveia<sup>1,3</sup>, Helena Freitas<sup>1,2</sup>, M. Teresa Gonçalves<sup>1</sup>, Rita Campos<sup>4,5</sup>

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Fungi are increasingly recognized as key players in ecosystem functioning, perhaps especially as decomposers, mutualists of plants, or as lichens. But fungi may be in trouble. Like animals and plants, fungi may be vulnerable to mass extinction. In recent years, fungal conservation has gained momentum. The need to consider fungi in conservation is increasingly accepted by the scientific community. However, if we want to bring fungal conservation to the fore, we must work to engage the public in a developing conservation narrative. Raising awareness of Fungi, emphasize how crucially important fungi are, and challenge attitudes about the fungi among the public is, therefore, paramount. In this communication, we introduce the concept of a "Celebration of Fungi Day", which aims to fight fungal blindness and change perceptions about fungi among the general public, celebrating their importance in nature and in our lives. We thought to do so by promoting a series of joint festive events to be held in a day of November of each year, in partnership with public and private institutions. We also share the experience that was the prefiguration of this concept, in the form of a one-day fungal festival that took place at the University of Coimbra, in November 2017. At that time, about 200 people participated in the various activities offered. We will present an overview of what we learned from this experience through the results of an online survey, complemented with free written comments by the public. Our findings suggest the value of delivering key messages about fungi during inclusive celebration events. The presentation will include a brief overview of the results of our participation in the Participatory Budget Portugal 2018, and highlight issues and perspectives for the future.

PS - poster session

**Comparative study of wood decaying fungi over a time line from Chakrata in northwest Himalaya in India**

Manoj Kumar, Nirmal S. K. Harsh, Anuradha Kumari  
*Forest Research Institute, Dehradun, India*

During a study conducted (2011-2014) in Chakrata region of northwest Himalaya in India, 164 wood decaying fungi were recorded. On reviewing the Fungarium records maintained at Forest Research Institute, Dehradun, India and publications, 125 species were found to be reported earlier from Chakrata (1922-1984). In the present study, 52 species recorded earlier could not be recorded again from this region and 43 new species were added to the list.

An attempt was made to prepare the threatened list of wood decaying fungi for this ecologically disturbed region. Though the time period and area was small to prepare such a list and did not strictly follow IUCN criteria, the present list has its importance in planning the conservation strategies for wood decaying fungi and to create awareness. The number of fungi was low and most of the species were represented by very small population (once or twice in 61 sampling units). Data on fungal species decline for this region was unavailable/ limited. Fungarium records have incomplete or absence of locality details and records of time series was almost lacking for the localities. Evaluation of decline was, therefore, performed indirectly through present status and available data from previous records. Information on habitat change and habitat decline was uncertain. An account of 217 species has been given in the list that included richness recorded in this study and earlier reported fungi, here 35 species were categorized under NT, 63 under LC, 68 under DD and 51 under NE category. Some more studies are needed to get a clear picture of fungal species status in this region. From this evaluation, 35 Nearly Threatened species require more attention and studies in the area following IUCN criteria.



PS - poster session

### **Evolution of the populations of ten macromycetes in Belgium since 1940**

**André Fraiture**

*Botanic Garden Meise, Meuse, Belgium*

The evolution of the populations of ten macromycetes has been studied in Belgium by analyzing a compilation of all the observations of these fungi made up to now in the country. These species have been selected for this study because they showed an expansion, sometimes so marked that it is almost possible to speak about “invasive species”. At least three different patterns of progression are observed, depending on the species.



PS - poster session

**Morphologically similar, cryptic taxa of macrolichens of conservation importance in the Carpathian Basin**

**Edit Farkas**<sup>1</sup>, László Lőkös<sup>2</sup>, Mónika Sinigla<sup>3</sup>, Nóra Varga<sup>1</sup>, Katalin Veres<sup>1</sup>

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Conservation of macrolichens has special importance, because most of them are conspicuous with considerable aesthetic value. If attention is directed to them being protected, superfluous collecting of any other lichens is restricted in general. Lichens serve as bioindicators in nature conservation, especially if indicate areas worth for protection. However, identification of their taxa is often difficult even for the specialists. Therefore it is important to find the adequate character sets and introduce further differentiating features and establish revised concept for segregating taxa.

Several genera contain morphologically similar species with different chemical compositions. Chemical revisions are especially important in case of lichens, where c. 1000 secondary substances exist and most of them are uniquely characterising lichens. High performance thin-layer chromatography is applied according to standard methods. Chemically revised genera often reveal species, which are rare at the investigated area or pointed out as new occurrences to a country's lichen flora. Analysing habitat features of these species, differences are shown in habitat preferences and ecological requirements.

These taxa are then suggested for protection, or their red list categories are established or modified. Examples are shown from genera *Cetrelia*, *Cetraria*, *Cladonia* (incl. *Cladina*), *Xanthoparmelia* (both usnic acid containing and brown species). Preliminary results from morphometric studies contra chemical analysis carried out in the species group *Cladonia chlorophaea* to analyse the value of morphological and chemical characters in identification process are outlined.

The work was supported by the project NKFI K 124341 financed by the National Research Development and Innovation Fund.



PS - poster session

### **Fungi in the Red Book of Belarus**

**Darya Belomesyatseva**<sup>1</sup>, Tatiana Shabashova<sup>1</sup>, Olga Gapienko<sup>1</sup>, Yadviga Shaporova<sup>2</sup>, Eugene Yurchenko<sup>3</sup>

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The Red List of the Republic of Belarus is a list of rare and threatening species of plants and animals. The Red List is the main state document on the basis of which scientific and practical measures directed to protection, reproduction and rational using of rare species have been developed. 25 species of lichens and 34 species of fungi were included in the last (4th) edition of the Red List (2015).

The list of fungi includes: Ascomycota: *Hydnotrya tulasnei* (Berk.) Berk. & Broome, *Ojibwaya perpulchra* B. Sutton, *Spathularia flavida* Pers., *Stephensia bombycina* (Vittad.) Tul. & C. Tul., *Tuber aestivum* Vittad., *T. borchii* Vittad. Basidiomycota: *Auriculariopsis albomellea* (Bondartsev) Kotl., *Bankera fuligineoalba* (J.C. Schmidt) Coker & Beers, *Boletopsis leucomelaena* (Pers.) Fayod, *Calvatia gigantea* (Batsch) Lloyd, *Cantharellus cinereus* (Pers.) Fr., *Clavariadelphus pistillaris* (L.) Donk, *Cortinarius cagei* Melot, *C. elegantissimus* Rob. Henry, *C. rheubarbarinus* Rob. Henry, *Dentipellis fragilis* (Pers.) Donk, *Fistulina hepatica* (Schaeff.) With., *Fomitopsis rosea* (Alb. & Schwein.) P. Karst., *Ganoderma lucidum* (Curtis) P. Karst., *Geastrum campestre* Morgan, *Grifola frondosa* (Dicks.) Gray, *Hericium coralloides* (Scop.) Pers., *Hygrocybe coccinea* (Schaeff.) P. Kumm., *Hygrophorus limacinus* (Scop.) Fr., *H. nemoreus* (Pers.) Fr., *Lepista sordida* (Schumach.) Singer, *Leucoagaricus nympharum* (Kalehbr.) Bon, *Polyporus umbellatus* (Pers.) Fr., *Pycnoporus cinnabarinus* (Jacq.:Fr.) P. Karst., *Rugosomyces ionides* (Bull.) Bon, *Scytinostroma odoratum* (Fr.) Donk, *Sistotrema raduloides* (P. Karst.) Donk, *Sparassis crispa* (Wulfen) Fr., *S. laminosa* Fr.

It should be noted that it was for the first time that a micromycete (*Ojibwaya perpulchra*) was included in the Red List.

PS - poster session

**Inoculation for conservation – developing protocols for inoculating wood-decay fungi into living trees for targeted conservation outcomes**

Matthew Wainhouse, Emma Gilmartin, Lynne Boddy  
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Modern forestry has moved away from coppicing and pollarding, to plantation forestry where whole trees are harvested at an earlier stage of growth. Under this practice, trees are removed before cavities and hollows can develop or dead wood form. This has been highly detrimental to woodland biodiversity as these micro-habitats have become increasingly rare in forest ecosystems. This issue is compounded by the fragmentation of woodland which prevents dispersal limited taxa, particularly fungi and invertebrates, from colonising new sites, even where management is favourable. Despite more sustainable approaches to forestry, there is a break in temporal continuity leaving a 'generation gap' between established old trees with hollows, deadwood and other microhabitats, and the young trees scheduled to replace them. Inoculating wood-decay fungi into trees and logs may be a useful conservation measure to bridge the gap. Specific fungi could be used to elicit a specific outcome, such as the creation of cavities or the establishment of a desired species.

Here we present two long-term field experiments where wood-decay fungi are inoculated into living trees as a targeted conservation intervention. The first aims to accelerate cavity formation in young beech trees (80 years old) by inoculating heart-rot species in large wood block inoculum. The second aims to reintroduce locally extinct populations of the rare wood-decay species *Hericium coralloides* and *H. erinaceus*, again through inoculation into young beech trees. We test different strains and substrates (sawdust and wood dowels) as inoculum. Through these experiments we aim to establish reliable and responsible protocols for using fungal inoculations as a targeted conservation tool.





PS - poster session

**An annotated checklist of rust fungi (Pucciniales) occurring in Portugal reveals endangered species, invasion routes and speciation hotspots**

**Rita Carvalho**<sup>1</sup>, Rui Figueira<sup>1,2</sup>, Ana Paula Ramos<sup>1,2</sup>, Pedro Talhinhos<sup>1,2</sup>

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The Pucciniales order comprise more than 5000 known species, arranged into 120 genera. A total of 2319 occurrences of rust fungi from 246 Pucciniales taxa were recorded in Portugal based on 115 publications and our own surveys, totalizing 683 rust taxon-host taxon unique combinations. This list was updated according to current taxonomic framework and georeferenced using the GBIF's recommended methods and associated with the vegetal host, providing a tool for diagnosis of rust fungi occurring in Portugal (and elsewhere in Europe), enabling to understand their spatio-temporal dynamics and to point out recent introduction of exotic species, along with situations of potential conservation value. Eleven species were recorded only in Portugal and are not reported for decades, raising great concern about their existence. Five species occur on endemic hosts whose populations are physically isolated from other hosts of the same rust species, mostly at the archipelagos of Azores and Madeira, illustrating scenarios where a speciation process may be occurring and thus urging the clarification of their taxonomic and ecological status. The geographical position of Azores and Madeira highlight these territories as entry/detection points for new rusts in Europe. Additionally, most of quarantine rusts currently absent from Europe are present in the American continent and several of them have suitable hosts in the Azores and Madeira. With a rust species:vascular flora ratio of 1:12.2, the Portuguese rust mycobiota can be considered well characterized. The proportion of number of species per genus/family in Portugal is similar to that found in other temperate countries, while tropical areas, and specially the neotropics, present the widest diversity.



PS - poster session

### **Mycology in Ireland - postcards from the edge**

**Maria Cullen**<sup>1</sup>, Howard Fox<sup>2</sup>

<sup>1</sup>*Barrow Herbarium, New Ross, Ireland*

<sup>2</sup>*National Botanic Gardens, Dublin, Ireland*

We discuss mycology in Ireland from Devonian times to our current work in understanding fungal species distributions and threats to fungi into the future.

The place of Irish fungi in law, in early scholarship, in the Irish language, and in contemporary use until the 17th century, is explored. Land ownership and land use change led to great modification of the Irish landscape after this time. Habitat destruction rather than industrial pollution was the main threat to Irish fungi in the 18th and 19th centuries. Since Irish independence in the early 20<sup>th</sup> century, the spread of *Hedera helix* (Ivy), air and water pollution, policy change, soil degradation, global supply chain impacts and lack of phytosanitary controls have all been factors in the decline of fungal species and their host trees. Our mycological work throughout the island of Ireland includes surveying for fungi in woods, plantation forests and fields, general lichen taxonomy and biodiversity research. To date, only one lichenised species, *Gyalolechia fulgens* (Sw.) Søchting, Frödén & Arup has been given legislative protection in Ireland as it has only been recorded at one site. Over 5,500 species of fungi have been recorded to date in Ireland. We propose a range of species for conservation toward a realistic effort to protect vulnerable fungi and their habitats in Ireland.

PS - poster session

**Diversity of macrofungi in the old growth spruce forest of the Stołowe Mts National Park,  
Poland**

**Izabela L. Kalucka**<sup>1</sup>, Andrzej M. Jagodziński<sup>2</sup>

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Fungi still belong to the most unexplored and least concerned components of biodiversity. The knowledge about the occurrence of fungal species, and in particular the actual data on the presence of rare, endangered and protected species in the area, is a prerequisite for undertaking effective actions aimed at protecting species and their biotopes as well as giving informed management recommendations.

Stołowe Mountains National Park (SMNP) belongs to the least studied Polish national parks in terms of knowledge of mycobiota (Kujawa 2017). A detailed inventory of macrofungi has been included in the series of research conducted in 2017 in the Czerwona Woda river valley (e.g., Dyderski et al. 2018, Wierzcholska et al. 2018). The observations were carried out in the close vicinity of the river bed along the distance of nearly 10 km within the SMNP, in the spruce old-growth forest planted in the potentially broadleaved area. Based on morphological examination of sporocarps and analyses of ITS region of nrDNA, nearly 230 species were identified. Among them are *Cortinarius fuscodiscus*, a species recorded for the first time in Europe; 17 species recorded for the first time in Poland (16 *Cortinarius* spp. and *Russula helodes*); 34 species from the Polish red list, as well as 2 species protected by law in the country, e.i. *Microglossum viride* and *Ganoderma lucidum*. *Cortinarius* was the most species-rich genus (32 taxa), with further collections which may represent species new for science. A century-long legacy of coniferous stands in PNGS has resulted in an impressive richness of fungi, especially of ectomycorrhizal group. However, an assemblage of old-growth wood-dwelling fungi is still relatively poor suggesting a long-lasting effect of forest continuity disturbance.

This study was supported by the Polish Forest Fund, General Directorate of State Forests, Warsaw, and by the funds for foreign research visits of the Faculty of Biology and Environmental Protection, University of Łódź.

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PS - poster session

### **Red list of the andorran fungi**

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*Centre d'Estudis de la Neu i de la Muntanya d'Andorra (CENMA)/Snow and Mountain Research Center of Andorra  
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Andorra is a mountainous country in the heart of the Pyrenees, with an altitudinal range from 848 to 2,942 m a.s.l. The vegetation are mainly forest of *Pinus sylvestris* and *Pinus uncinata* and alpine and subalpine meadows. The predominant habitat is the forest of *Pinus uncinata* with *Rhododendron ferrugineum*. In the bottom of the Valley predominates the Mediterranean vegetation with forest of *Quercus* and different kind of secondary vegetation product of the abandonment of traditional agricultural activities.

Our objective was to identify those species of threatened macromycetes of the Andorran mycoflora. Although, until now, there are about 600 of macromycetes known, and the list is increasing every year. The actual study of the threatened mushrooms of Andorra was made from a previous list of about 500 species. The degree of threat was established based on the guidelines of the IUCN. Because the size of the territory (468 km<sup>2</sup>), we used the regional analysis (IUCN, 2012).

The intensive sampling of mushrooms in Andorra started after year 2000, so, until now, we cannot infer any population dynamic in our study. For this reason, an evaluation of the populations is proposed, analyzing the data collected from the neighboring territories (France and Spain) and their red lists of fungi.

The results of our study are: in the category of Critically Endangered (CR) no species has been detected; Endangered (EN) 7 species, which is 1.39% of the total; Vulnerable (VU) 23 (4.58% of the total); Near Threatened (NT) 30 (5.97%); 392 species fits in the category of Least Concern (LC) that are 78.08%. In the category of Data Deficient (DD) and Not Evaluated (NE) there are 44 species (8.76%). Finally, there are 6 species (1.19%), whose presence has not been confirmed.

It has been found that many of the species in the highest threatened categories are part of the mycoflora of the snowbeds, associated with *Salix herbacea*, mainly because the effect of the climate changing.

PS - poster session

**Preliminary analysis of the first year of data on the study of the diversity and production of mushrooms in black pine forests (*Pinus uncinata*)**

Manel Niell

*Centre d'Estudis de la Neu i de la Muntanya d'Andorra (CENMA)/Snow and Mountain Research Center of Andorra, Institut d'Estudis Andorrans (IEA) - Andorran Research Institute, Andorra*

The objective of our study is measure the production, Phenology and diversity of macromycetes in *Pinus uncinata* forests.

We have installed seven plots of 10 x 10 meters placed at different heights and orientations. The plots have been sampled once a week during mid-July until the first snow, which this year 2018, took place at the end of October. We collected all the samples of mature specimens.

The samples have been determined to the level of species when it has been possible, measured the fresh weight, dried and finally, measured the dry weight. The number of species found is about approximately 110 (the correct identification of problematic species of some taxonomic groups such as *Cortinarius* and *Inocybe* is still pending). We found 1315 mushrooms that correspond to 2359.32 gr. of dry weight.

The variation among plots has been large enough, even for those plots that were one near the other. Jaccard's diversity index, for example, between the two most similar species, located about 50 meters from each other and with a slight change in orientation, is 0.25. Indexes of 0.10 and 0.11 are observed in plots located at different heights and orientations.

The vast majority of species, more than 60%, have only been located on a plot. The only species cited in five plots was *Lactarius quieticolor*. Present in 4 plots was *Clitocybe gibba*, *Paxillus involutus*, *Russula xerampelina*, *Tricholoma portentosum*.

The average production of mushroom per species was 11.53 and more than 50% of the species made between 1-10 carpophores. The most productive were: *Russula xerampelina* (15% of total weight); *Tricholoma portentosum* (14% of total weight) and *Russula integra* (16.6% of total weight). The *Cantharellus cibarius* appears in 2 plots.

The season 2018 was very rich in species and in production of mushrooms because the abundant precipitations during the month of July, August and September. In mid October, the end of the rains and the wind interrupted the apparition of mushrooms. In the end of October, the first snow, ended with the mushroom season.

## DATA SESSION



OS - oral session

**The formal and informal sources of data on protected and red-listed species in Poland**

**Małgorzata Ruszkiewicz-Michalska**<sup>1,2</sup>, Julia Pawłowska<sup>3</sup>, Anna Kujawa<sup>1</sup>

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The good-quality data on distribution and population conditions of protected, red-listed and rare fungal taxa are necessary for the assessment of actual level of threat, as well as the planning and adjusting of protection measures. Poland, as other European countries, is obliged by national and international regulations to collect data about protected fungal species and to assess fungal biodiversity in order to prevent its losses (eg. via monitoring of rare taxa). Formally, this duty relies on diverse national institutions, including landscape and national parks, state forests as well as General and Regional Directorates of Environmental Protection. However, in Poland, there is no central and comprehensive database on protected, threaten or rare fungal species. There is a number of small databases launched by both law-obligated institutions and groups of amateur mycologists.

In order to gather information on distribution of taxa protected in Poland all these institutions were questioned. The data turned out to be fragmentary, scattered, not thoroughly reliable, and mostly inaccessible. The gap of an institutional database of valuable species is successively filled by the publicly available, informal register of protected and endangered fungi (GREJ; [grzyby.pl/rejestr-grzybow-chronionych-i-zagrozonych.htm](http://grzyby.pl/rejestr-grzybow-chronionych-i-zagrozonych.htm)) that was launched in 2005. This database is the result of cooperation between professional mycologists and advanced amateurs. In addition to protected species, it also collects data on species that are new, rare or insufficiently sampled in Poland.

There is an urgent need to build a nationwide system of collecting mycological data (not only on species currently protected and endangered) and to establish rules for making its resources publicly available.

OS - oral session

**PacBio sequences from material samples are incorporated into UNITE Species Hypotheses datasets**

**Urmas Kõljalg**

*University of Tartu, Natural History Museum, Tartu, Estonia*

UNITE (<https://unite.ut.ee>) is a community of mycologists established in 2001. Our community includes scientists as well as Citizen Scientists. The ambition of UNITE is to develop datasets and digital tools for robust and reproducible molecular identification. We also developed Digital Object Identifiers (DOIs) based system for the communicating fungal species. Datasets of the nuclear ribosomal internal transcribed spacer (ITS) region, form the basis for UNITE. The current version includes nearly 1 million public fungal ITS sequences. Datasets are curated and annotated by community members. During the past 15 years, they made nearly 300 000 improvements. In the absence of Latin names for fungal species, UNITE offers a unique communication system where species hypotheses (SH) are provided with Digital Object Identifiers (DOIs).

The current version 8 of UNITE offers more than 800 000 DOI-based SHs. These DOI identifiers are also incorporated into the taxonomic backbone, making communication of taxa seamless in both directions. DOI identifiers of species hypotheses are also used by GBIF (Global Biodiversity Information Facility) in order to publish high-throughput sequencing taxon occurrence data.

UNITE serves as a data provider for a range of metabarcoding software pipelines like QIIME, mother, CREST, UCHIME, etc. Recent improvements include ITS-based species hypotheses for all eukaryotes which helps to place non-fungal ITS sequences in accurate taxon.

The most challenging enhancement of the UNITE in 2019 was aggregation of full-length, high-quality ITS sequences generated by the PacBio Sequel system from soil and other material samples.



OS - oral session

**Establishing GBIF infrastructure in Poland: opportunities and challenges**

Piotr Tykarski

*Department of Ecology, Biological and Chemical Research Centre, Faculty of Biology, University of Warsaw, Warsaw, Poland*

Global Biodiversity Information Facility<sup>i</sup> is a leading global initiative aimed at making species data available for multi-purpose use. Having achieved the stage of its development, on which immense data volumes are mobilised, GBIF introduced biodiversity information into the realm of “big data”. Currently, its distributed database system offers more than 1,328 million of occurrence records, published from more than 45,000 datasets, by 1,420 institutions. The network attracts more and more attention from scientific community every year, with more than 3,700 peer-reviewed papers using GBIF data.

The specific position of GBIF as a scientific multi-purpose tool comes from the fact that it uses information technologies to describe biodiversity and relationships of its multifaceted aspects to various fields and disciplines, standing strongly on precise scientific terminology and evidence. The data published by GBIF members are interoperable, discoverable and re-usable which vastly increases its usability. Thus, a particular specimen of a xylotrophic fungus, a voucher to an occurrence record shared in GBIF, may be used for an analysis of distribution of a certain taxonomic group of medical importance, or in an article on a possible shift in boreal species ranges due to climate change, in a report about an species inventory for a conservation plan of a forest, or in a historical paper describing heritage of a famous mycologist who collected and deposited the polypore, in the same time.

The network is both a technical infrastructure and an international organization, having a governance structure, long-term strategy, work plans and partner organizations. The organization relies almost entirely on contributions of participant countries. Typically the progress of GBIF technologies reflects advances in biodiversity informatics in countries who contribute the most to the organization. This is not, however, the case of Poland whose government made a decision to become a full GBIF member only in 2017, after 16 years of participating at a non-binding level of the associated membership.

GBIF's goals and ideas are being implemented in Poland by the Polish Biodiversity Information Network<sup>ii</sup> (Krajowa Sieć Informacji o Bioróżnorodności, KSIB). Although the local network has been contributing data to the global infrastructure (more than 1,5 million occurrence records in 92 databases published by 25 member institutions), the level of development and maturity of the infrastructure at a country scale should be seen as a basic one. Despite of it, KSIB created a number of projects, with the most prominent, long-term Biodiversity Map<sup>iii</sup> – an integrative approach to data on species (currently insects and spiders). It seems that the most needed requisite for the local network is the substantial investment to create and organize a robust technical infrastructure for biodiversity data that would be closely linked to scientific institutions and governmental bodies. There are, however, alternative paths that can bring us closer to the goal - the easily available meaningful information on biodiversity.

In the most advanced countries of the GBIF family, the infrastructure and its technologies constantly increases its importance in the local biodiversity-related disciplines and applications among many audiences, e.g. scientific community thanks to improvements of publishing policies

or amateur naturalists through partnerships with citizen science initiatives. In some cases GBIF is in use for national monitoring and biodiversity reporting purposes. It is very important to understand the catalytic role for facilitating access to biodiversity information that GBIF-related actions can play in Poland and other countries. Thanks to standardized data schemas, the global network may and should become a generic ultimate platform for different groups of data publishers who otherwise would not cooperate with each other and the data they produce would remain scattered.

OS - oral session

**Fungi, sequences, and global biodiversity: boosting the impact of mycology through primary data**

**Dmitry Schigel**

*GBIF - Global Biodiversity Information Facility, Copenhagen, Denmark*

GBIF—the Global Biodiversity Information Facility—is an international network and research infrastructure funded by the world’s governments and aimed at providing anyone, anywhere, free and open access to data about all types of life on Earth, including multifaceted data on Fungi. Coordinated through its Secretariat in Copenhagen, the GBIF network of participating countries and organizations, working through participant nodes, provides data-holding institutions around the world with common standards and open-source tools that enable them to share information about where and when species have been recorded. The GBIF network draws together data from disparate sources like museum collections, field surveys and citizen scientists’ observations—more than 1.3 billion species occurrence records in total. More than 1,400 institutions worldwide share datasets under machine-readable Creative Commons designations, which supports its use and application by scientists, researchers and others in hundreds of peer-reviewed publications and policy papers each year. Many of these analyses—which cover topics from the impacts of climate change and the spread of invasive and alien pests to priorities for conservation and protected areas, food security and human health— would not be possible without this big-data resource.

As the world’s largest collection of spatiotemporal evidence for life on Earth, GBIForg provides a central access and discovery point for these data. Its heterogeneity leads to in the intrinsic taxonomic, spatial and other biases, not unlike the biases in the literature in the largest libraries. As with published resources, digital biodiversity data resources require data postprocessing and filtering to ensure optimal data fitness for use in research and practice. Adequate data use is of course only possible if data content is digitally and centrally available. GBIF works as an overarching system with general search and filter mechanism by a few common parameters, such as species name, locality, date and a few others. Once a data resource is shared into the GBIF index, data can be downloaded or accessed through GBIF API. Advanced users can also use these platforms as discovery mechanisms for exploring data at source, where they can access higher levels of detail on parameters not indexed by GBIF, such as sequence alignment, host, associated organisms, etc. In this way GBIF supports both data access and data discovery while coexisting with the thematic and niche data systems.

By 12 June 2019, fungal data in GBIF are represented by 16.7 million occurrence records and over 217 thousand names (Fig.1), dominated by human observations and preserved specimens (Fig. 2).

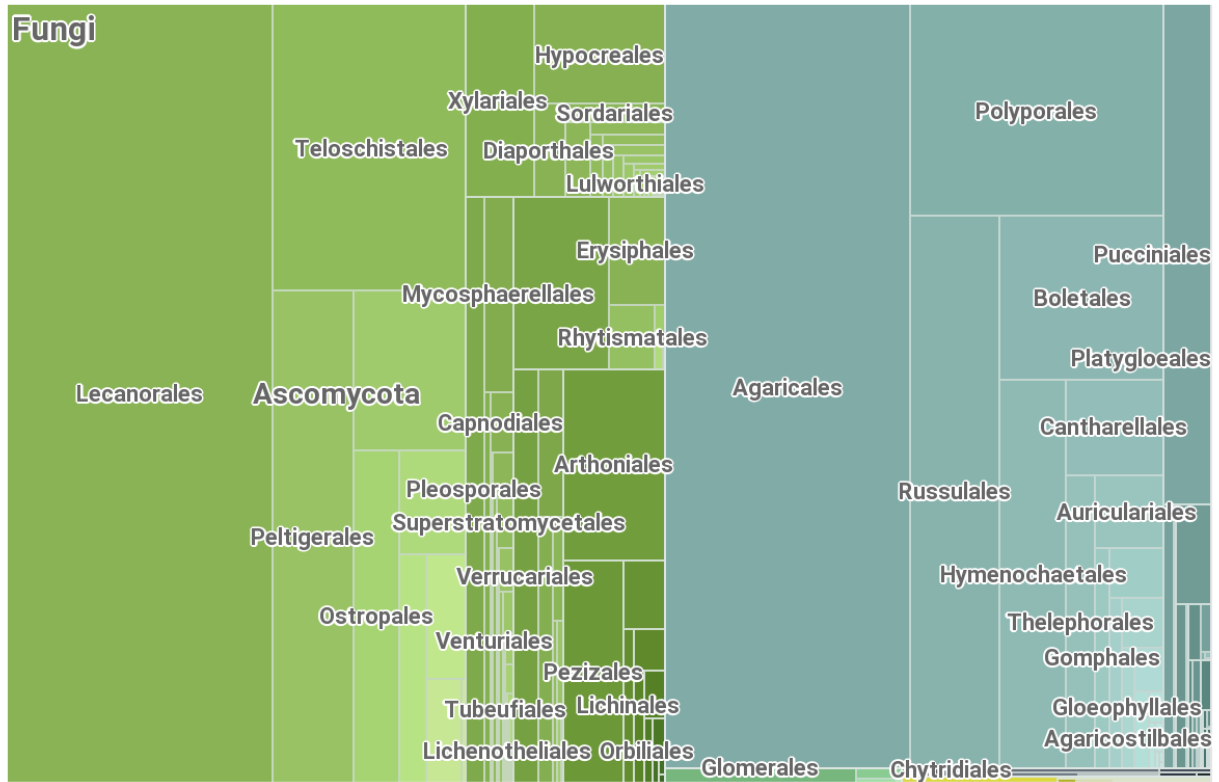


Fig. 1. Taxonomic distribution of fungal occurrences in GBIF [www.gbif.org/species/5](http://www.gbif.org/species/5) by 12 June 2019.

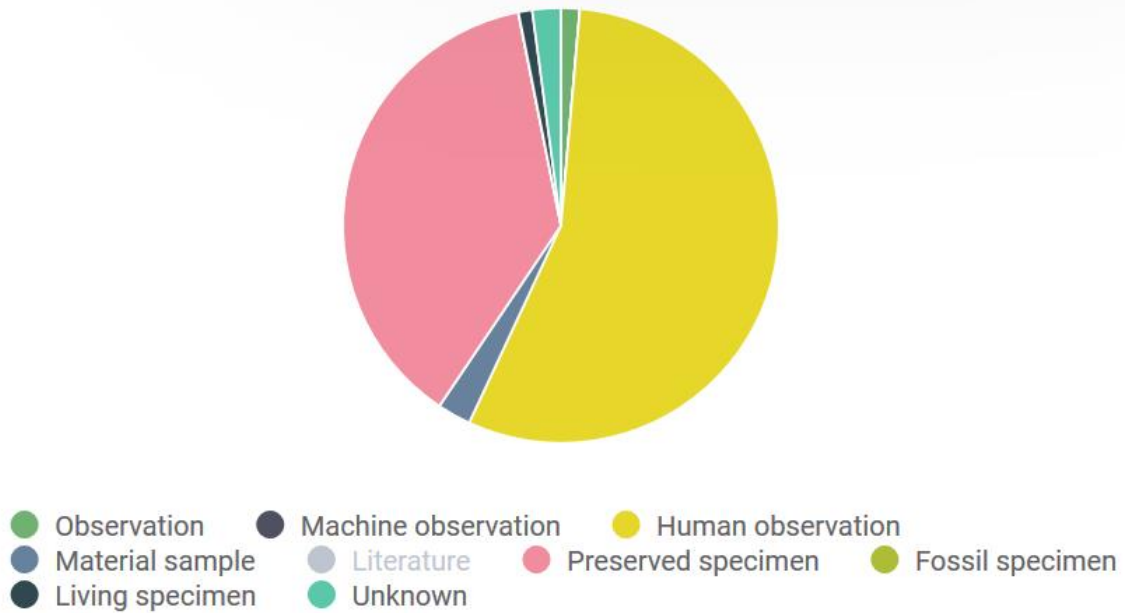


Fig. 2. Number of fungal occurrences with different bases of record in GBIF [www.gbif.org/species/](http://www.gbif.org/species/), by 12 June 2019.

Molecular data, in particular metagenomics, have a huge potential to rapidly combat taxonomic bias in GBIF-mediated content, and since 2018 GBIF has worked with UNITE (Estonia), European Bioinformatics Institute (UK), and the International Barcode of Life to add new Linnaean and OTU names to the GBIF taxonomic backbone, enabling GBIF to index the growing data stream of morphologically and molecularly detected biodiversity. The presentation will cover the status and future plans of this work and highlight individual, organisational and national benefits of integrated data approach in mycology.

## OFFERED PRESENTATIONS

OS - oral session

**Fungal alkaline proteases and enzymes with activity of blood proteins**

**Alexander Kurakov**<sup>1</sup>, Alexander Osmolovskiy<sup>1</sup>, Anna Bobrovskaya<sup>1</sup>, Yulia Pokrovskaya<sup>1</sup>, Yakov Dunaevsky<sup>2</sup>  
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There is a demand for alkaline proteases and the proteases with the activity of proteins of the hemostatic system. For the selection of producers of these enzymes studied range of new species and strains of alkalophilic and alkalotolerant fungi (*Sodiomyces alkalinus*, *S. magadii*, *Chordomyces antarcticum*, *Acrostalagmus luteoalbus*, *Verticillium zaregamsianum*, *Gibellulopsis nigrescens*), entomopathogens and nematophagous species (*Beauveria bassiana*, *Purpureocillium lilacinum*, *Paecilomyces carneus*, *T. cylindrosporum*, *T. inflatum*), and saprotrophs - *Aspergillus sclerotiorum*, *A. nidulans* and *A. ochraceus*. Proteolytic activity was determined in the culture liquid by azokazein, collagenolytic activity – by azocoll, fibrinogenolytic activity - by fibrinogen, specific proteinase activity – by synthetic peptide para-nitroanilide substrates for endopeptidases and exopeptidases, substrates of Xa-factor of blood plasma, plasmin, thrombin, activated protein C, tissue plasminogen activator. The strains with high fibrinolytic activity and activator for protein C were found. Optimized conditions for their cultivation, primary purification of enzymes and characterized their properties were done. One of the strains has shown the indirect fibrinolytic action due to activation of plasminogen and protein C-like activity. High protease activity in conditions close to the blood suggests their effective action in vivo. The strains with the highest total proteolytic, metalloproteases, subtilization, dipeptidyl-peptidases and aminopeptidases activities under alkaline conditions were identified among alkalophiles and alkalotolerants. The activity of cysteine proteinases, rare for fungi, was found. The strains with high activity of proteases at pH 6-10, increased content of NaCl, Triton X-100, SDS and detergent Ariel, H<sub>2</sub>O<sub>2</sub> and liquid stain remover Vanish Oxi Action were selected. It shows the possibility of their practical application.

OS - oral session

**Effective usage of edible mushrooms extract for treatment of HeLa cells**

**Inesa Avagyan**<sup>1,3</sup>, Siranush Nanagulyan<sup>3</sup>, Liya Minasbekyan<sup>2</sup>

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Mushrooms are considered as functional foods because they elicit their positive effect on human being in several ways. Stimulation of host immune defense systems by bioactive polymers from medicinal mushrooms has significant effects on the maturation, differentiation and proliferation of many kinds of immune cells in the host. Many of these mushrooms polymers were reported previously to have immunotherapeutic properties by facilitating growth inhibition and destruction of tumor cells.

Early we have been determined the anti-inflammatory activity of such three wood-decaying mushrooms culture as *Pleurotus ostreatus* P. Kumm., *Ganoderma lucidum* (Curtis) P. Karst and *Lentinula edodes* (Berk.) Pegler, which during growth has treated by mm-waves of different frequency. The treatment of mushrooms led to modulation in their enzymatic activity. Highest activity among these three mushrooms has expressed the *P. ostreatus*, whose peroxidase activity increases in 3 times, and  $\beta$ -glucosidase -2 times. Intraperitoneal injection of the extract obtained from this culture suppresses an acute inflammation by 85% on the model of rat ear acute inflammation, induced by xylol. The proteins of extracts was separated in PAAG and studied by HPLC. Revealing candidate compounds, which has such extraordinary activity.

The purpose of this work is to estimate the anticancer activity of artificial activating cultures extract of *P. ostreatus* on the continuous cell line of HeLa cells in vitro. As evidence our data, extract from *P. ostreatus* markedly suppressed growth of tumor human cultures: has a suppressive action to 48h of cultivation and almost completely suppressed mitotic activity of HeLa cell line cultures. Nuclear morphologic changes associated with the loss of differentiation, growth inhibition, damage of the cell membrane till the destruction of tumor cells. We will discuss the antitumor activity of obtained by us extracts of *P. ostreatus* and suggest candidate compound in the content of extract having a protein nature.



OS - oral session

**Filamentous fungi and yeast associates of phoretic mites on *Ips typographus* in Eastern Finland**

**Riikka Linnakoski**<sup>1</sup>, Ilmeini Lasarov<sup>2</sup>, Pyry Veteli<sup>1</sup>, Olli-Pekka Tikkanen<sup>2</sup>, Risto Kasanen<sup>3</sup>, Heli Viiri<sup>1</sup>, Tuula Jyske<sup>1</sup>, Tuan A. Duong<sup>4</sup>, Michael J. Wingfield<sup>4</sup>

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The European spruce bark beetle (*Ips typographus*) has become a major forest pest in Finland in recent years. The intervals between outbreaks have become shorter and thus the damaging effects more significant. The beetle is a well-known vector of mainly ophiostomatoid fungi causing blue-stain of wood, and pathogens that have the ability to amplify the insect damage. It also carries other organisms, such as phoretic mites. The ecology of these mites is poorly understood. In this study we examined fungi associated with mites phoretic on *I. typographus*. Adult bark beetles were collected from wind-blown spruce trees (*Picea abies*) in Joensuu, Eastern Finland during June-August 2017. A total of 180 beetles were inspected for the presence of phoretic mites. In total, 39 phoretic mites identified as *Uropodina species* (Acari: Mesostigmata) were found, of which seven were nymphs. The mites and nymphs were removed from the beetles, and fungal associates were isolated from them. Fungal identifications were based on DNA sequences and phylogenetic analyses of the ITS and  $\beta$ -tubulin gene regions. Fifteen fungal species were detected, including eight yeasts and seven filamentous fungi. Eleven percent of the mites carried fungi, and of these 74% carried at least one fungal species. An average of two fungal species were carried per mite. The most commonly found filamentous fungi were *Grosmannia penicillata* (25 %), *Ophiostoma bicolor* (19 %), *O. ainoae* (12 %) and *O. brunneolum* (12 %). The most commonly found yeast was *Wickerhamomyces bisporus* (47 %). The other yeasts included *Chionosphaera cuniculicola* (Basidiomycota), and two species of *Ogataea* and *Kuraishia*, as well a putatively novel species residing in genus *Nakazawaea* (Ascomycota).

OS - oral session

**Ectomycorrhiza of *Tricholoma vaccinum*-spruce: Interactions and metal tolerance**

**Katrin Krause**<sup>1</sup>, Katharina Wagner<sup>1</sup>, Wilhelm Boland<sup>2</sup>, Erika Kothe<sup>1</sup>  
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<sup>2</sup>Max Planck Institute for Chemical Ecology, Jena, Germany

Diverse organisms interact in the ectomycorrhizospheric habitat, including the host plant, mycorrhizal fungi, and other rhizosphere microorganisms. Different signaling molecules may influence the ectomycorrhizal symbiosis.

In a forest biotope, the mutual symbiosis between the basidiomycete *Tricholoma vaccinum* and Norway spruce (*Picea abies*) was characterized by a high diversity in basidiomycetes and a rich bacterial community. The bacterial isolates showed symbiosis-relevant traits with 74 % producing the phytohormone indole-3-acetic acid, 23 % producing siderophores, and 23 % mobilizing phosphate.

The mycorrhizal fungus *T. vaccinum* was able to excrete plant hormones into the medium upon axenic cultivation. These include auxins, salicylic and abscisic acid, and jasmonates. The spruce roots exudated auxins and salicylic acid. With these compounds present in soil of a natural ectomycorrhizospheric habitat, a communication network is present. A response of *T. vaccinum* to the environmentally available salicylic and abscisic acids led to altered hyphal branching relevant for mycorrhization.

RNA-Seq analysis comparing fungal pure culture with mycorrhizal co-cultures was performed to examine the molecular biological background of the interaction. Using a treatment with seepage water from a former uranium mine additional effects of metal and salt contamination on mycorrhizal interaction were investigated. Thus, new insights from the finely tuned phytohormone interactions in the mycorrhizosphere are presented showing a specifically rich system to study microbial communication.

OS - oral session

**Mycobiota of the soil contaminated by copper molybdenum mining**

**Ruzanna Matevosyan**<sup>1</sup>, Vahagn Gevorgyan<sup>2</sup>, Tigran Yesayan<sup>1</sup>

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Fungi have an important role in the nature. They can be found everywhere but majority of them live in soil. Samples of soils were taken near the territory of Zangezur copper molybdenum combine (Armenia) and its environs. For mycological preliminary analysis of micromycetes there was used dilution method which is based on the use of the water-soil suspension and its transposition on the agar medium (pH = 4.5, to prevent the growth of bacteria). The cultivation of fungi was carried out at temperature 23-25°C. Cultivation duration was from 4 to 14 days. As a result of soil investigation 20 species of fungi were detected, 3 of which belong to the Zygomycota (*Mucor genevensis*, *M. genevensis*, *Rhizopus stolonifer*), 16 species belong to the Ascomycota (*Alternaria alternata*, *Aspergillus candidus*, *A. clavatus*, *A. flavus*, *A. fumigatus*, *A. niger*, *A. ochraceus*, *A. versicolor*, *Fusarium solani*, *Penicillium canesceas*, *P. cyclopium*, *P. lanosum*, *P. multicolor*, *Trichoderma koningii*, *Verticillium glaucum*, *V. heterocladium*), and 1 belong to the Basidiomycota classes (*Sclerotium oryzae*). Systematics is given according to modern classification. The received data indicate that the changes in the composition of the species of micromycetes of soils and their structural changes can be used as indicators of air pollution and for studying the degree of the anthropogenic impact on various ecosystems.

OS - oral session

**Contents of ergothioneine in fruiting-body of wild strain *Lentinula edodes* collected in Korea**

**Rhim Ryoo**, Hyo-rim Lee, Yeun Sug Jeong, Yeongseon Jang, Kang-Hyeon Ka  
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*Lentinula edodes* called shiitake is an edible and medicinal mushroom that has been popular for Eastern Asia as well as Europe. *L. edodes* was often found in temperate forest at over 1,000 m above sea level and at the stems and stumps of Oak tree (*Quercus* spp.). Various new varieties of *L. edodes* have been developed cautiously with the increasing interest in health. This mushroom has been used as healthy functional food because of low in calories, rich in proteins and chitin, various vitamins. Some research have been reported concerning ergothioneine of various new strain *L. edodes*. Ergothioneine is an effective antioxidant and a cellular protector against oxidative activity. In study, content of ergothioneine measured using the fruiting-body wild strain *L. edodes* collected in Korea and then compared their content with several new varieties using commercially available mushrooms. To analyze the ergothioneine content of *L. edodes*, freeze-dried mushroom powder(1g) was added to 20 ml extracting solution. After 4 ml of 10g/L sodium dodecyl sulphate solution was added and the mixture was centrifuged at 25°C and 2,000 rpm. HPLC condition consisted of 10µL sample loop, inno C-18 column(4.6x250 mm). The mobile phase was 0.5M phosphate in 30% acetonitrile and 1 ml/1L trimethylamine adjusted to a pH of 7.3. Ergothioneine was quantified by the calibration curve of the authentic standard. The results showed that amount content of ergothioneine have a differ according to the type of mushroom strains.

OS - oral session

**Forest management's alteration of the underground fungal biodiversity in  
Mediterranean coniferous forest**

**Segula Masaphy**

*MIGAL and Tel Hai College, Kiryat Shmona, Israel*

The underground composition of the fungal community plays an important role in forest plant productivity and health as well as to the mushroom productivity as a byproduct of the forest. Forest management actions may alter both above mushroom diversity as well as the underground general soil mycobiome. We have studied the effect of different thinning regimes on the underground fungal community in LTR station in a coniferous forest in Israel 9 years after thinning. The fungal community composition, species abundance and richness, were dramatically changed according to the thinning regime. Moreover, alteration of the fungal community according to the different ecological groups (saprotroph, pathogens, mycorrhizal) was also observed in relation to the thinning regime.

OS - oral session

**Space for time: Soil fungal communities and biomass across a boreal forest age gradient**

**Luis Morgado**<sup>1</sup>, Sunil Mundra<sup>1</sup>, Ella Thoen<sup>1</sup>, O. Janne Kjønås<sup>2</sup>, Yngvild Ransedokken<sup>3</sup>, Line Nybakken<sup>3</sup>, Erik Stange<sup>4</sup>, Olav Skarpaas<sup>5</sup>, Björn Nordén<sup>4</sup>, Håvard Kausrud<sup>1</sup>

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Boreal forests are shaped by modern forestry practices, including clear-cutting, leading to a mosaic of various even-aged forest stands. In this study, we aimed to reveal how soil fungal communities in boreal forests changes with stand age. Fungal communities were analyzed in 61 forest plots dominated by spruce, pine and birch, 2 to 250 years old. Fungal communities were investigated using ITS2 DNA metabarcoding while fungal biomass was estimated using ergosterol. Climatic, vegetation and soil edaphic variables were also recorded. Although an extensive list of explanatory factors were examined, most of the variation in fungal community composition was left unexplained. Soil edaphic factors such as soil pH, C content and soil moisture accounted for more of the compositional variation than forest stand age. On the other hand, fungal biomass correlated strongly with forest age, being highest in old pine forest stands. The proportion of ectomycorrhizal fungi was higher in younger stands with a higher coverage of spruce, while saprotrophs showed higher relative abundant in forest stands with acidic soils and higher C content. Taken together, our results indicate that the composition of soil fungal communities in boreal forest soils are mainly governed by soil edaphic factors and not to forest age per se while fungal biomass is more strongly related to forest age.

OS - oral session

**Biodiversity and news records of genus *Chaetomium* in Egypt**

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We characterized the genus *Chaetomium* associated with desert, salt marshes soil, seaweeds, herbivore dung, medicinal plants, and archaeological wood samples from Egypt. Here we summaries more than three years extensive data on genus *Chaetomium* in Egypt. Twenty-one species were isolated from the different ecological habitats and were studied for genetic diversity based on the phylogenetic analyses in comparison to morphological data on genera and species level. According to this study, *Ch. globosum* is the most abundant Chaetomiaceae in Egypt (28/66), followed by *Ch. iranianum* (5/66) and *Ch. grande* (4/66), *Ch. madrasense*, *Ch. murorum* and *Ch. subaffine* (3/66) each and the abundance of the remaining taxa either one or two out of 66 isolates. The morphological diversity of ascospores, texture, appendages of the new records were illustrated by light and scanning electron microscope. It is intended, in the present taxonomic study, considered a revision of the generic concept of *Chaetomium* in Egypt and provides a new insight into the phylogenetic relationships among different taxa within *Chaetomium*. The specimens were deposited at the Fungarium of Suez Canal University (Egypt). A key to the 21 *Chaetomium* species recorded from Egypt is also provided.

OS - oral session

**New data on phytopathogenic Ascomycota of Ukrainian Polissya**

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Phytopathogenic ascomycetous fungi of the northern part of Western Polyssia were studied during last years in Ukraine for the first time. Natural vegetation of the area has mostly migratory character and boreal plant species with Holarctic and Eurasian range are dominant. The fraction of plant species on their range limits is about eight hundred species that provides favourable conditions for phytopathogenic fungi development. Observations during summer period revealed that among leaf invading anamorphic Ascomycota the species from the order Capnodiales were dominant, and the group of fungi from Mycosphaerellaceae family (33 species) was the largest. Less abundant were fungi of the order Pleosporales (15 species) and the other orders of Amphisphaeriales, Botryosphaeriales, Glomerellales and Helotiales numbered 1-5 species. As a result of survey new for Polissya fungi from the genera *Neoramularia*, *Pestalozziella*, *Ramularia*, *Septoria* were registered. Interesting species of *Septoria hydrocotyles* was observed for the first time in Ukraine. The fungus is known in Western Europe, Asia, Australia and North America. It caused abundant leaf lesions on rare marshy plant *Hydrocotyle vulgaris*. The rare fungus of *Neoramularia bidentis*, that was described from the South Korea and recently registered in Poland, was collected on *Bidens tripartita* for the first time in Ukraine also.

The list of species specifies fungi of the other genera as well: *Ascochyta*, *Caryophylloseptoria*, *Cercospora*, *Cladosporium*, *Colletotrichum*, *Dendryphiella*, *Discosia*, *Epicoccum*, *Venturia*, *Gloeosporidiella*, *Microsphaeropsis*, *Monilia*, *Phoma*, *Phyllachora*, *Phyllosticta*, *Pseudocercospora*, *Septogloeum*, *Sphaerulina*, *Stachylidium*, *Thyrostroma*, *Torula*.



OS - oral session

### **Confusion in the Taxonomy of genus *Phellinus***

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With the advent of molecular analytical tools status of many fungal groups has been revised. Molecular methods are considered to resolve relatedness problems and help to create more natural grouping of fungal species rather it is causing more chaos and confusion due to lack of clear cut guidelines. Effect of this chaos can be seen in case of genus *Phellinus* established in 1886 by Quélet. Initially this genus was considered monophyletic but Murrill in 1907 split *Phellinus* into smaller genera but and in 1984 Fission and Niemela reassessed European species of *Phellinus* using morphological, numerical, chemical and cultural studies and introduced two new genera. With the introduction of molecular techniques many workers reassessed genus *Phellinus* which made it clear that this genus is a polyphyletic group and splitted it into subgenera and genera under *Phellinus* sensu lato. With molecular phylogenetic studies, nomenclature and grouping of genus *Phellinus* has constantly been in chaos. Examples of *Phellinus pini* (Brot.) Pilát established as type specimen of group *Porodaedalea* and named as *Porodaedalea pini* (Brot.) Murrill again reverted to *Phellinus pini*; *Phellinus linteus* (Berk. & M.A. Curtis) named as *Fulvifomes linteus* (Berk. & M.A. Curtis) Murrill, *Pyropolyporus linteus* (Berk. & M.A. Curtis) Murrill, *Inonotus linteus* (Berk. & M.A. Curtis) Teixeira and to *Tropicoporus linteus* (Berk. & M.A. Curtis) L.W. Zhou & Y.C. Dai and *Phellinus robiniae* (Murrill) A. Ames confirmed as species of *Fulvifomes* but again placed back to *Phellinus*. This constant nomenclature and group status change creates confusion among taxonomists and researchers. It is high time that guidelines and regulations be put in place for changing species names and groups to minimize such confusion and chaos.

OS - oral session

**A molecular phylogenetic assessment of the genus *Scutellinia* (Pezizales: Pyronemataceae)**

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Pyronemataceae is the largest and most heterogeneous family of Pezizales, encompassing 78 genera and approximately 660 currently recognized species.

*Scutellinia* spp. is a cup fungus belonging to the Pyronemataceae (Ascomycota). Searching the Species fungorum database for the genus *Scutellinia*, 258 records were found and among these 125 were currently considered valid *Scutellinia* species.

This cosmopolitan genus is easily recognized by its shield-like red or orange apothecia, globose to ellipsoidal ascospores with or without various wall ornamentations, and stiff, long, blackish-brown hairs that arise from inner cells of the ectal excipulum. Among the characters that have previously been used in defining species, hair morphology and ascospore shape and ornamentation have been especially emphasized in most keys at the infrageneric and species level (Schumacher 1979, 1990).

Despite considerable taxonomic study of the genus, species delimitation and infrageneric subdivision within *Scutellinia* have remained problematic. Molecular phylogenetic approaches have been applied at the species level to other members of the Pyronemataceae, but only a few *Scutellinia* species have been included in larger level studies (Perry et al. 2007).

In this study the phylogenetic relationships among a large set of *Scutellinia* spp. (20/25) were explored based on the comparison of the sequences of the internal-transcribed spacer regions ITS1 and ITS2 (including the 5.8S rRNA gene). This was the first time that this fungal genus was subjected to a molecular phylogenetic analysis. The results were compared with morphology-based classification schemes, with the objective of evaluating the phylogenetic significance of characters such as hair morphology, ascospore shape and ornamentation, and habitats.

PS - poster session

**Oral candidiasis in hiv infected patients and its antifungal susceptibility pattern by disc diffusion method**

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Oral Candidiasis (OC), being a sign of immune imbalance that usually heralds the transition to AIDS, is the most common manifestation in patients infected with HIV. The low absolute CD4+ T-lymphocyte count has traditionally been cited as the greatest risk factor for the development of OC and current guidelines suggest increased risk once CD4+T-lymphocyte counts fall below 200 cells/mm<sup>3</sup>. Rampant, indiscriminate and long-term use of antifungals has led to the development of antifungal resistance among *Candida* species. Hence, a cross-sectional study was conducted in Sukraraj Tropical and Infectious Disease Hospital (STIDH), Teku, Kathmandu.

A total of 408 oral swab samples were collected from patients with and without active lesion visiting ART (Anti-Retroviral Therapy) center of STIDH. Sample was processed according to standard microbiological guidelines.

*Candida albicans* (n=53) was the most common isolated organism followed by *Candida tropicalis* (n=3), *Candida krusei* (n=2), *Candida glabrata* (n=1), and other *Candida* species (n=6). Among the 65 yeast isolates from HIV seropositive individuals, 26 patients had CD4+ ≤ 200 cells/mm<sup>3</sup> and 39 patients had CD4+ >200 cells/mm<sup>3</sup>. 14 patients exhibited severe immune suppression with CD4+ count less than 100 cells/mm<sup>3</sup>. There was a significant association between few cases like: oral *Candida* carriage and CD4+ cell count ≤ 200 cells/mm<sup>3</sup> (p<0.001), URTI and oral *Candida* isolation (p=0.029), recent antibiotic consumption and oral *Candida* isolation (p=0.002). In contrary, there was no significant association between ART use and oral *Candida* infections (p=0.188), no association between tobacco consumption and Oral *Candida* isolation (p=0.051).

PS - poster session

**Evaluation of Three Mushroom Species Ethanol Extracts in the Treatment of Prostate Cancer in Wistar Albino Rats and Phytochemical Analysis of the Fungi in Nigeria**

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Globally, prostate cancer is the sixth leading cause of cancer-related death in men. In the United States it is the second. Prostate cancer is most common in the developed world with increasing rates in the developing world. However, many men with prostate cancer never have symptoms, undergo no therapy, and eventually die of other unrelated causes. Many factors, including genetics and diet, have been implicated in the development of prostate cancer. The presence of prostate cancer may be indicated by symptoms, physical examination, prostate-specific antigen (PSA), or biopsy. Prostate-specific antigen testing increases cancer detection but does not decrease mortality. The purpose of this study is to evaluate the antiprostate cancer activity of mushroom extract in treatment of prostate cancer in wistar albino rats. Prostate cancer was induced experimentally on the animals by administering intraperitoneally with Estradiol and Testosterone solution for 21 days. The prostrate level was monitored by sacrificing one of the normal rats and compared with the control group in other to know the effect of the extract. The effect of the herbal medicine on the prostate specific antigens (PSA) compared to the controls. There was significant difference in the prostate cancer animal treated with herbal medicine compared to the normal control and control groups. Also, no significant decrease was observed in the control groups. The result of the phytomedicine and Fenasteride effect on the biochemical parameters. The haemoglobin level of the prostate cancer rat treated with the drug and Fenasteride were significantly high compared to induce untreated. There was a clear indication that the extract preparation could be safe for use. The study showed that the extract preparation had some antiprostate cancer activity on the prostate specific antigens and also on other parameters examined. The study also revealed that the drug doses investigated did not provoke toxic effect to the animals.

PS - poster session

**Characterization of an extracellular laccases and decolorization of synthetic dyes from mycelium of *Lentinula edodes* (shiitake)**

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*Lentinula edodes* (Shiitake) is an important edible mushroom in the world. *L. edodes* is distributed in Asia and some subtropical regions and is a white rot fungus with excellent wood-degrading ability. The lignin activity of this species can be measured using laccases, which are multi-copper oxidase that catalyze the transformation of aromatic and non-aromatic compounds with reduction of molecular oxygen to water. Synthetic dyes are phenolic compounds which can be used to measure laccase activity. In this study, we investigated the correlation between the growth of strains and laccase activity by using the decolorization of different synthetic dyes and measure vitality according to the storage period. Mycelial growth, chromaticity change, dye decolorization and laccase activity were measured using six synthetic dyes for ten strains of this species. In the color of media containing methyl red, mycelial growth was slow and chromaticity change was weak. However, in the color of media containing bromocresol purple, bromothymol blue and congo red, the chromaticity change shows high and has higher correlation than other dyes. The correlation coefficient among dye decolorization, mycelial growth, and laccase activity was not so significant. In the color media using bromothymol blue, decolorization ratio and laccase activity showed higher correlation than other dyes. In preserved strains, the longer stored media showed the less mycelial growths and a low laccase activities. These data suggest that these processes are considered to be an useful methods for measuring the viability of fungal strains.



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