

**7TH CZECH-SLOVAK MYCOLOGICAL CONFERENCE,
PRAGUE, SEPTEMBER 16–18, 2021****ENGLISH VERSIONS OF ABSTRACTS (ELECTRONIC SUPPLEMENT)**

The seventh Czech-Slovak Mycological Conference took place in Prague from 16th to 18th September. It was organised by the Czech Scientific Society for Mycology, the Slovak Mycological Society and the Institute of Microbiology of the Czech Academy of Sciences, whose lecture hall in the campus in Praha-Krč was the venue of the whole event (except the Saturday excursion to the forests above the Sázava river valley). Of the 74 registered participants, 63 mycologists were present personally (52 from Czechia, 10 from Slovakia and 1 from Austria), while the remaining 11 were connected online. In two cases also the lectures were presented online.

Complete abstracts of all lectures and posters in Czech are published in the printed version of Mykologické listy. Enjoy the reading.

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Monika Kolényová et al.: Microhabitats of lignicolous fungi

Karel Švec et al.: Study of endosymbionts of the bark beetle *Ips typographus* (in Czech only)

Barbora Křížková et al.: Taxonomically related, yet ecologically contrasting bark beetle species host fundamentally distinct microbial communities

SESSION 4 – FUNGAL SYSTEMATICS I

Petr Hrouda: Notes from Czech Mycology

Michal Tomšovský et al.: Phylogeny and species delimitation of *Hymenochaete* in the Czech Republic

Vladimír Antonín et al.: European species of the *Melanoleuca castaneofusca* group

Jan Holec et al.: Cleaning the Augean stables in the group of *Gymnopilus stabilis*, *G. sapineus* and *G. penetrans* (in Czech only)

Viktor Kučera: Does *Glutinoglossum glutinosum* grow in Slovakia? – News on the research into earth tongues (Ascomycota, Geoglossaceae)

SESSION 5 – FUNGAL SYSTEMATICS AND DIVERSITY II

Roman Labuda et al.: Ten new fungal species described at the BiMM research platform (Tulln, Austria) during 2018–2021

František Sklenář, Vít Hubka: Species delimitation in *Aspergillus*: revision of section *Flavipedes* and more

Luboš Zelený: Macromycetes of Diana Nature Reserve, Český les Mts. (in Czech only)

Aleš Jirsa: Tomentelloid fungi occurring in South Bohemia

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Markéta Šandová: Specimens of the genus *Hypomyces* in the PRM herbarium

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Jan Holec, Juan Carlos Zamora: *Ditiola haasii* (Dacrymycetaceae), a rarity from Boubínský prales (in Czech only)

Daniel Dvořák: Earthstar surprise from Podyjí National Park (in Czech only)

SESSION 6 – DIVERSITY AND IDENTIFICATION OF FUNGI

Tomáš Větrovský et al.: GlobalFungi – a global database of fungal occurrences

Miroslav Kolařík: Ribosomal DNA – better the devil you know

Ondřej Koukol, Greg Delgado: Practical (and tragic) consequences of identification of fungi based on DNA barcodes only

Jan Borovička et al.: Cadmium hyperaccumulation in *Thelephora penicillata* and its isotopic composition

Tereza Veselská et al.: Application of flow cytometry in the study of ecology and functional traits of lichens

Lukáš Janošík et al.: Ascospore morphology of bryophilous Pezizales is closely associated with host ecology and position of infection

INVASIVE OOMYCETES IN FOREST NURSERIES AND THEIR MANAGEMENT

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Species of the genus *Phytophthora* belong to the most important pathogens of forest trees, and their introduction into forest stands (where they are most often introduced with planting material) is the cause of serious damage. Research carried out in dozens of forest nurseries of the country in 2017–2020 confirmed the considerable extent of infestation of growing areas and materials. A total of 44 oomycete taxa were detected, of which 22 belonged to the genus *Phytophthora*. The most commonly identified alien or cryptogenic invasive species were *P. plurivora*,

P. gonapodyides and *P. cambivora*, but extremely dangerous alien species such as *P. citrophthora* and *P. cinnamomi* were also identified.

In reality, the problem of contamination probably affects all or the vast majority of nurseries (with a total area of about 1500 ha), where about 210 million forest plant seedlings were produced in 2019. Based on the results, a qualified estimate of dozens of millions (probably more than 100 million) forest seedlings are contaminated annually. It is disturbing fact that one of the most sensitive trees to oomycetes is beech, with a production of 65 million seedlings, now used as the main tree species in the reforestation of calamity clearings. Under the project, a methodology for integrated protection of forest planting material against *Phytophthora* pathogens was developed in order to create and maintain hygienically appropriate nurseries and to produce safe nursery material to be used broadly in artificial restoration without fear. The methodology is based on a new proactive approach to the problem and is divided into three sections dealing with disease symptomatology, preventive and curative measures. The key part is preventive measures, without which the production of healthy material cannot be applied consistently.

The work was supported by project TA ČR TH02030722.

WHAT IS NEW IN THE GENUS *PHYTOPHTHORA* (PERONOSPORALES, OOMYCETES, STRAMENOPILA)?

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Phytophthora is a serious plant pathogen. Many *Phytophthora* species cause extensive damage to woody plants, a reason why this genus is the subject of a large international research project at Mendel University in Brno. The principal aims of the project are an investigation of global and local *Phytophthora* biodiversity, investi-

gation of host-pathogen interactions, the description of new species and testing the impacts of such new, insufficiently known *Phytophthora* species from tropical areas on European native tree hosts.

One of the most important results of the project is the discovery of the probable origin of *Phytophthora ramorum* in the Laurosilva forests of East and Southeast Asia (Japan and Vietnam). Attention was also paid to the influence of increased CO₂ concentrations on the development of *Quercus robur* disease by simultaneous infection of the root pathogen *P. plurivora* and consumption of leaves by *Lymantria dispar* caterpillars. Another study described the interaction between *P. cinnamomi* and sweet chestnut (*Castanea sativa*) during pathogen infection. We surveyed the effects of *P. cinnamomi* infestation on stem tissues immediately bordering the infection site compared to more distant parts of the trunk. Infection by *P. cinnamomi* led to massive reprogramming of the chestnut proteome and accumulation of stress hormones (salicylic and jasmonic acids). We confirmed that inoculation experiments linked with proteomic and metabolomic methods lead to the identification of unknown molecules involved in the pathogenicity of *P. cinnamomi*.

Besides *Phytophthora* monitoring in forest and ornamental nurseries, different methods of *Phytophthora* isolation from soil and water were tested to evaluate frequency and diversity of *Phytophthora* species at the local level.

The work was supported by the *Phytophthora* Research Centre project, reg. no. CZ.02.1.01/0.0/0.0/15_003/0000453.

PREDICTING POTENTIAL IMPACT OF INVASIVE ALIEN PATHOGENS ON WOODY VEGETATION IN CZECH NATURA 2000 HABITATS

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The number of invasive alien tree pathogens as well as the risk of severe damage to host trees and decline in their population have been increasing globally in recent decades. This can lead to changes in the structure and species composition of invaded ecosystems, impairment of their functions and threats to biodiversity. Predictive

modelling is increasingly being used as a useful management tool to control invasions and protect host tree populations, forest ecosystems and the landscape as a whole.

We chose eleven pathogens with the actually or potentially highest impact on Czech nature, including four *Phytophthora* species (*P. alni*, *P. cinnamomi*, *P. plurivora* and *P. ramorum*), two pathogens of maple trees (*Eutypella parasitica* and *Cryptostroma corticale*), *Hymenoscyphus fraxineus* and *Dothistroma septosporum*. We used geographic information systems with expert knowledge species distribution models to develop spatial predictions of i) the environmental suitability for these pathogens, and ii) their potential impact on woody vegetation in Natura 2000 habitats. Climatic, topographic and soil characteristics as well as data on the presence of watercourses and the occurrence of host trees were used as predictors.

The resulting maps show large areas potentially at high risk of invasion by the studied pathogens. They also demonstrate that locations, landscape types as well as Natura 2000 habitats vary in susceptibility to invasion and a combined effect of different invasive pathogens can be expected in many of them. We believe that the developed models and maps can help to implement effective management strategies to monitor and control invasions of these organisms in the Czech Republic and minimise their impact on the landscape.

This work was supported by the Technology Agency of the Czech Republic under project TH03030306.

ALIEN FUNGAL PLANT PATHOGENS IN THE CZECH REPUBLIC

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The issue of the spread of alien organisms has been a worldwide highlighted topic for over 30 years. During this time, many lists and databases of alien organisms documenting the distribution of mainly higher plants and animals have been created. However, the spread of microorganisms is somewhat delayed due to their hidden way of life and difficult identification. Based on a request by the Czech Ministry of Agriculture, the aim of our work was to create a list of alien fungal and fungal-like plant pathogens detected in our country after the year 2000, in the first stage focusing

on pathogens of woody and ornamental plants. The occurrence of 500 alien species known in Europe was confirmed in Czechia based on browsing databases, literature sources and herbarium specimens. Of pathogens of woody plants and ornamental plants, about 80 new species have been introduced in the past 20 years. Another 120 as yet unrecorded species known in Europe may already occur in Czechia or will be introduced in the near future. About half of these species are polyphagous and at risk of rather easy domestication and causing greater damage to ecosystems. A total of 40 species were assessed as the most hazardous, most often representatives of the classes Oomycetes and Sordariomycetes. Alien fungal pathogens spread to Czechia mostly from neighbouring countries and from countries with a similar climate. The original distribution ranges of these species are very often unknown. For species with a known native range, North American and Asian origins predominate, which is related to the main trade partners in Europe.

This study was supported by the Scientific Committee on Phytosanitary and Environment and the Center for Landscape and Biodiversity (DivLand) under TA ČR (Technology Agency of the Czech Republic) project SS02030018.

***PLASMOPARA HALSTEDII* ON SUNFLOWER IN THE CZECH REPUBLIC**

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Since 2007, sunflower downy mildew has been recorded at 10 localities in the Czech Republic. The isolates of its causal agent, *Plasmopara halstedii* (Farl.) Berl. & De Toni, were studied in pathotests, i.e. the evaluation of phenotypic reactions of 15 differential sunflower lines. Presence of the following races (named according to their virulence code) was revealed: 70060, 70471, 70571, 71060, 71461, 71471, and 71571. All Czech isolates have been found to be sensitive to metalaxyl. Superinfection by the *Plasmopara halstedii* virus was found in 73% of Czech isolates in a recent screening (RT-PCR, specific primers, electrophoresis) carried out in 2014–2020. Occurrence and frequency of individual races in *Ph* populations is continuously developing in many countries. Genetic analysis of European *Ph* populations using polymorphic

microsatellite markers showed differences in the Czech populations which share alleles with populations of *Ph* from several other countries.

Acknowledgement: IGA UP PŘF-2021-01, the Czech National Programme on Conservation of Microorganisms and Invertebrates of Agricultural Importance.

**PATHOGENICITY OF TWO RELATED SPECIES,
DOTHISTROMA SEPTOSPORUM AND *D. PINI***

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Dothistroma needle blight (DNB) is an important fungal disease caused by two species of *Dothistroma*: *Dothistroma septosporum*, which occurs worldwide, and *D. pini*, which has a restricted distribution. Differences in susceptibility to the disease have been observed in many pine species. The aim of this study was to compare the virulence of *D. septosporum* and *D. pini* and to find out the differences in susceptibility to *Pinus nigra* and *P. mugo*. In 2019, two-year-old pine seedlings were planted in two naturally infected stands with DNB (Kálnica experimental area infected by *D. septosporum* and Jahodná infected by *D. pini*). The seedlings were exposed to natural infectious pressure. The seedlings were cut down after 2 years. Needles were collected and classified into three categories: (1) healthy needles without damage, (2) chlorotic needles, and (3) needles with lesions caused by *Dothistroma*. At Kálnica, the seedlings of both tree species were damaged at the same intensity: the proportion of needles with lesions caused by *D. septosporum* was 42.4% for *P. mugo* and 40.5% for *P. nigra*. At Jahodná, infected by *D. pini*, *P. nigra* seedlings were damaged more (23.3%) than *P. mugo* seedlings (12.9%). This suggests that *P. nigra* is more susceptible to *D. pini* than *P. mugo*. Plants infected by *D. septosporum* were more severely damaged than those infected by *D. pini*, suggesting that *D. septosporum* is more virulent than *D. pini*. No symptoms of the DNB disease were present in the control seedlings. This preliminary result needs to be confirmed and supplemented by other more detailed studies (e.g. higher number of replicates and experimental plots in order to exclude the influence of climatic and habitat conditions, more detailed evaluation of the infection intensity of infested needles).

The study was financially supported by project VEGA 2/0077/18 and bilateral project APVV SK-FR2017-0025.

SUSCEPTIBILITY OF *PINUS ARMANDII* TO *DOTHISTROMA SEPTOSPORUM*

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The fungus *Dothistroma septosporum*, causing Dothistroma needle blight (also known as red band needle blight) is one of the most serious pathogens of pine trees. It has not only a wide geographical distribution, but also a wide host range. The host spectrum of *D. septosporum* (confirmed by molecular identification methods based on DNA) includes 55 taxa of 6 genera (*Abies*, *Cedrus*, *Larix*, *Picea*, *Pinus* and *Pseudotsuga*), the main host being *Pinus*. *Pinus armandii* is missing from the list of host species of this fungus. The aim of this study was to determine the susceptibility of this pine species to *D. septosporum*. In the experiment, we used 2–3 year old *P. armandii* seedlings, from which we removed all damaged needles. For artificial inoculation we used a spore suspension of the fungus *D. septosporum* M621 (*P. mugo*, Bzenica SR, 23.2.2017) with a concentration of 3×10^6 in 1 ml of distilled water. Using a multi-purpose hand sprayer, we equally sprayed 15 ml of spore suspension around the whole seedling and incubated the plants in a cultivation chamber at 95% humidity with a temperature and light regime of 16 h light at 20 °C and 8 h dark at 12 °C. As a positive control we used *P. nigra* seedlings and as a negative control *P. armandii* plants sprayed with distilled water. The pathogenicity test will be evaluated 12–14 weeks after the artificial infection.

The first preliminary results of visual evaluation suggest that *P. armandii* is susceptible to *D. septosporum*, indicated by the first necrotic spots on the needles recorded 8 weeks after inoculation, which are typical of Dothistroma needle blight.

This work was supported by research project VEGA 2/0077/18.

ZOOPHILIC DERMATOPHYTES ASSOCIATED WITH PET ANIMALS

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Dermatomycoses are skin and skin adnexal diseases of warm-blooded animals and one of the most common human infections. The agents of these diseases are members of a group of fungi from the order Onygenales called dermatophytes. The spectrum of dermatophytes isolated from European patients has only slightly changed in the past few decades. The only exceptions are zoophilic species, especially from the *Trichophyton benhamiae* complex, which shows stronger fluctuations in incidence. With the mechanisation of agriculture and introduction of vaccination programmes, dermatomycoses transmitted from livestock animals to humans have decreased while dermatomycoses transmitted from pet animals have significantly increased. The increment is mainly related to the introduction of new pathogens of the complex from non-European countries to Europe and to the increasing popularity of breeding exotic animals, e.g. hedgehogs.

The project was supported by the Czech Ministry of Health (grant AZV 17-31269A), Charles University Research Centre programme (no. 204069) and a long-term research project of the Academy of Sciences of the Czech Republic (RVO: 61388971).

INACTIVATION OF DERMATOPHYTES CAUSING ONYCHOMYCOSES USING NON-THERMAL PLASMA AS A PREREQUISITE FOR THERAPY

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Onychomycoses or fungal diseases of the nails are a neglected problem in the human population. The available preparations often have side effects or are not sufficiently effective. Non-thermal plasma (NTP) is an ionised gas with disinfectant effects. It can inhibit the growth of microorganisms and has therefore the potential to support the treatment of onychomycosis. An apparatus for the generation and application of NTP has been designed for experiments and for therapy. Four types of dermatophyte micromycetes were selected based on their frequency of occurrence and severity of effects to patients in clinical practice. To compare the efficacy of NTP on micromycetes across species and strains, a minimum of two strain representatives were selected for each species. To compile a therapy proposal, a matrix of NTP exposures was determined and the experiments were divided into 2 groups, namely NTP exposures in early stages of micromycete growth and exposures in advanced stages of development. The experiments show that the differences in susceptibility of micromycetes to NTP are not only found at the interspecies level, but also within individual strains. Exposures of micromycetes in early stages of growth are effective, leading to inhibition or inactivation of micromycetes. Conversely, in the case of already developed micromycetes, they can only lead to growth retardation but not to complete inactivation. Therefore, NTP therapy in combination with nail hygiene is recommended as removal of visibly grown fungi on the nail plate or nail bed supports exposure in early stages of micromycete development. To support the therapy, it is advisable to disinfect the environment, such as the patient's shoes, to prevent relapses and recurrence. The designed equipment for NTP generation is also suitable for disinfecting the surroundings.

The project was partially funded by research programme Progress Q25 of Charles University Prague.

HOSPITAL-RELATED MICROSCOPIC FUNGI AND THEIR RESISTANCE TO DISINFECTANTS

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Fungal pathogens causing many kinds of infections are increasingly being identified as agents related to hospital-required diseases, esp. in immunocompromised patients. Airborne fungal propagule load may vary strongly between different departments of even the same hospital, usually during warm and humid months. The aim of the study was to analyse comprehensively hospital air mycobiota, specially focused on wards with the highest hygienic standards, followed by a characterisation of the relationship between hospital air-related mycobiota and employed disinfection regimen as well as an analysis of the sensitivity of fungal isolates to disinfectants applied in hospitals. All fungal isolates were identified according to their macro- and micromorphology. The selected representatives were tested for sensitivity to disinfectants based on polyhexamethylene guanidin (PHMG), quaternary ammonium salt or peracetic acid according to the STN EN 1650 protocol. Pathogenic (*Aspergillus fumigatus*), toxic fungi (*A. versicolor*, *Fusarium* spp. etc.), and clinically severe opportunistic pathogens (zygomycetes, *A. terreus*) were recovered from the hospital indoor atmosphere. This finding is highly alarming from the perspective of hospital hygiene. The PHMG did not show any antifungal activity (according to the latest data of the European Agency for Chemicals, the compound has not been rated as a biocide anymore since this summer). The disinfectant based on peracetic acid showed the highest antifungal efficacy. In accordance with our expectations, zygomycetes, melanised fungi and fungi producing macroconidia were more resistant to the disinfectants.

MICROFUNGAL OCCURRENCE IN AIR OF SPELEOTHERAPEUTICAL SPACES

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Breathing inside caves (speleotherapy) is used to treat pulmonary diseases with children as well as adult persons, such as bronchial asthma, allergic colds, sino-bronchitis, dermorespiratory syndrome, and recurrent catarrhs of the upper respiratory tract, currently also to treat post-covid patients. Speleotherapy for children has been practised in the Czech Republic for more than 40 years (Císařská Cave, Sloupsko-Šošůvka Caves, and Zlaté hory) and speleotherapy spaces for adult persons are in preparation, too.

The occurrence of airborne microfungi was studied inside speleotherapy spaces in several caves of the Czech Republic, Slovakia and Hungary (Čísařská, Sloup-Šošůvka, Domic, Jasovská, Baradla, and Béke). Isolation of airborne spores was also carried out in adjoining cave spaces and in air outside caves. In the process, colony-forming units (CFU) as well as the spectrum of microfungi were compared.

AIRBORNE FUNGAL SPORES IN PRAGUE

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Spores of microscopic filamentous fungi, together with pollen grains, are airborne allergens, which can cause allergic reactions in sensitive individuals at higher concentrations.

The aim of the study was to develop a methodology for the sampling and identification of microscopic fungi in the air and in indoor environments, usable for monitoring health-threatening allergenic fungal species. At the same time, the risk of various localities in Prague was evaluated.

Sampling took place at 3 locations from the beginning of May to the end of October using a cordless handheld vacuum cleaner. A Petri dish with potato-carrot agar was placed in the vacuum cleaner.

Species of the genus *Cladosporium* were the most abundant, accounting for 90–92% of all isolated microscopic fungi at the localities. Species of the genus *Alternaria* were recorded with a frequency of 2–4%. Species of the genera *Botrytis*, *Epicoccum*, *Penicillium* were also present.

During the sampling of aeromicrobes in the outdoor environment from April to October, it was found that the main peak occurs at the turn of July and August. Another small peak was detected at the end of September. These peaks with the highest incidence were found repeatedly in both monitored years 2019 and 2020. The indoor environment of flats was dominated by similar genera of fungi as in the outdoor air. Only the proportion of representatives of the genus *Cladosporium* was lower and, conversely, the genus *Alternaria* showed a higher proportion.

A total of 109 isolates of the genus *Alternaria* were obtained from the outdoor air. Their genus identification was confirmed by PCR with primers Dir5cAlta1 and

Inv4Alta1. Most isolates could be classified as *Alternaria alternata* based on PCR results with primers AaltDAlta1 and AinflAlta1. The presence of the gene for Alt a 1, the major allergen of *A. alternata*, was confirmed in most isolates using primers Alta1CF and Alta1CR.

The results were obtained with the support of the Operational Programme Prague – Growth Pole of the Czech Republic, reg. no. CZ.07.1.02/0.0/0.0/17_049/0000830.

SUCCESSION OF FUNGAL COMMUNITIES IN FORMERLY MINED RAISED BOGS IN THE ŠUMAVA MTS.

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Restoration of industrially mined raised bogs is a long-term and difficult process, dependent not only on abiotic conditions, but also on the possibilities of species dispersal from the surroundings. Its success is usually evaluated based on vegetation analysis and the presence of tyrphobionts (species strictly associated with peatlands). Although fungi play an important role as plant symbionts, endophytes, pathogens, or decomposers of dead biomass, they have not yet been studied in detail during the restoration of raised bogs. Our aim was to find out whether fungal succession in restored peat bogs (Soumarský Most and Vlčí Jámy) corresponds to the vegetation succession and how fungal communities differ from those of native *Pinus rotundata* forests (Malá Niva and Velká Niva).

In the first year of the research, 241 fungal species forming fruitbodies were recorded and 217 operational taxonomic units (OTUs) were identified from peat and decaying *Sphagnum* by means of environmental sequencing in 24 permanent plots (9 × 9 m). Both methods are able to distinguish succession stages of fungal communities, which correspond to the vegetation. Fruitbody data better reflect vegetation because they contain specialised saprotrophs associated with aboveground parts of plants. By contrast, environmental sequencing detects micromycetes and fungi forming ericoid mycorrhizae, describing the underground diversity better. Surprisingly, the 15–25 year old succession stages of restored peat bogs come closest

to the communities of *Pinus rotundata* forests. Older stages (25–35 years) are transformed into birch-dominated stands with significantly different mycobiota. Tyrophobionts occur most frequently in *Pinus rotundata* forests and also in the youngest stages (up to 15 years), where they grow on bare peat and in *Eriophorum vaginatum* bults.

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MICROHABITATS OF LIGNICOLOUS FUNGI

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We studied the diversity and ecology of lignicolous fungi on dead wood in natural forests. We described the microhabitat preferences of individual species and morphological groups on decaying beech logs using data collected in 2015 from four nature reserves (Polom, Kohoutov, Žofínský prales, Žákova hora). We analysed the microhabitat preferences (wood decay, vertical/horizontal position, cover of bryophytes/bark in area of fructification, growth on specific habitats – woody material fallen from a log, hollow, surface breakage) of species and morphological groups.

Species were found to prefer a specific position on logs, which is partly related to fruitbody morphology. The strongest pattern was apparent in the vertical position of fruitbodies on logs. Fully resupinate species were frequently found at the bottom of the log while pileate species (agarics, polypores) at the top. Pyrenomycetes and perennial polypores fructify on less decayed wood. On the other hand, fleshy fungi (agarics) prefer more decayed parts of logs (including woody material fallen from decaying logs).

**TAXONOMICALLY RELATED,
YET ECOLOGICALLY CONTRASTING BARK BEETLE SPECIES
HOST FUNDAMENTALLY DISTINCT MICROBIAL COMMUNITIES**

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The microbial communities associated with bark beetles are shaped by complex abiotic and biotic variables. We examined fungal associates of bark beetles infesting European Ash (*Fraxinus excelsior*), namely *Hylesinus fraxini*, attacking thin branches, and *H. crenatus*, boring in trunks of fully grown trees. Material of different ontogenetic stages of both beetle species and their galleries was examined using cultivation and metabarcoding approach focused on composition and abundance of fungi. Although both beetles live on the same host tree and are closely related, they host clearly distinct fungal communities. Thin branch borer *H. fraxini* relies mostly on *Geosmithia* (Ascomycota: Hypocreales) symbionts, whereas the trunk-infesting *H. crenatus* is predominantly associated with *Ophiostoma hylesini* (Ascomycota: Ophiostomatales). This trend is also visible in other bark beetle species on other trees for which we have metabarcoding data only. We hypothesise that simple abiotic variables related to the microhabitat of each beetle species such as moisture, water stability and CO₂ content in larval galleries are responsible for the differences in communities of fungal symbionts. Surprisingly, a large fraction of the cultivated symbionts are undescribed species, mostly yeasts from both phyla (Ascomycota, Basidiomycota).

NOTES FROM CZECH MYCOLOGY

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Does Czech Mycology look more like an exotic than a Czech journal, at least in some issues? Does it mean that it is a world-famous journal or, as some people argue, that we give too much space to exotic authors? Let's look at a few examples, which 'exotics' sometimes submit papers to our journal.

For many of them, indexing in the Scopus database is attractive. It is also appreciated by our authors, at least in fields of applied science. As errors can occasionally occur in large databases, even here it is sometimes necessary to fine-tune some details.

Finally a look inside the editorial process. Four eyes see more than two, but how many eyes are needed to check really everything to secure that the article will not be published with a serious mistake? And what happens when you use the help of an online translator?

PHYLOGENY AND SPECIES DELIMITATION OF *HYMENOCHAETE* IN THE CZECH REPUBLIC

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The phylogeny of the genus *Hymenochaete* (Hymenochaetales, Basidiomycota) has not yet been sufficiently investigated in Europe. The aim of the work was to clarify the relationships of the recently described species *H. jaapii* and *H. pilatii* (formerly *Hymenochaete tabacina* f. *crocata*, reported as missing in the Czech Republic) to other local *Hymenochaete* species (*H. carpatica*, *H. cinnamomea*, *H. cruenta*, *H. fuliginosa*, *H. rubiginosa*, *H. subfuliginosa* and *H. ulmicola*) and the sister genus *Hydnoporia* (*H. tabacina* and *H. corrugata*). The results of DNA sequences of the ITS region of the ribosomal RNA gene and translational elongation factor 1-alpha revealed that *H. jaapii* and *H. pilatii* are well distinguished from other European species; *H. pilatii* was rediscovered at localities in the surroundings of Prague and close to

Litomyšl. We also confirm a new record of *H. ulmicola* in the Czech Republic (Habrová Seč Nature Reserve SW of Moravské Budějovice, growing on a standing tree of *Ulmus glabra*, leg. J. Běřák). This species was historically described by A. Pilát as *H. rubiginosa* f. *minuta* and only the type specimen from 1912 has been recorded (leg. R. Picbauer, locality of Brodek near Olomouc). The current distribution of *H. ulmicola* in the Czech Republic is unknown.

The sequences of *H. fuliginosa* revealed interesting results. While the DNA sequences of *H. fuliginosa* collections from spruce (*Picea abies*) are easily distinguishable from those of *H. subfuliginosa* from oak (*Quercus* spp.), the sequences of most *H. fuliginosa* collections from fir (*Abies alba*) are almost identical to those of *H. subfuliginosa* (one collection from fir belonging to the *H. fuliginosa* spruce genotype). The phylogenetic differentiation of the *H. fuliginosa* fir genotype from *H. subfuliginosa* was not statistically significant.

EUROPEAN SPECIES OF THE *MELANOLEUCA CASTANEOFUSCA* GROUP

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Melanoleuca belongs to the taxonomically most difficult genera of agarics with several phylogenetic lineages. Species with either urticoid or missing cheilocystidia are classified into subgen. *Urticocystis*. European species of the *M. castaneofusca* group (*M. fontenlae*, *M. acystidiata*, *M. castaneofusca*, *M. stridula*, *M. luteolosperma*, *M. microcephala* and *M. paedida*) are summarised in this lecture. *Melanoleuca acystidiata* and *M. fontenlae* were described as new species this year. The study was based on morphological and molecular data of either type material or recently collected specimens from various parts of Europe. All species are briefly characterised

by macro- and micromorphological features and a multigene phylogenetic analysis of a combined dataset (ITS, rpb2 and tef1). In concordance with the results, a new taxonomical reassessment of *M. pseudopaedida*, *M. pseudoluscina* (both conspecific with *M. luteosperma*) and *M. robertiana* (nomen confusum) was proposed. The specimens previously identified as *M. robertiana* were described as *M. acystidiata*. *Melanoleuca pseudopaedida* as conceived by Vizzini et al. (2011) is different from the type of the species and belongs to *M. fontenlae*. The differences between the type material of *M. angelesiana* from the USA and collections identified as *M. angelesiana* from Europe are also discussed.

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**DOES *GLUTINOGLOSSUM GLUTINOSUM* GROW IN SLOVAKIA? –
NEWS ON THE RESEARCH INTO EARTH TONGUES
(ASCOMYCOTA, GEOGLOSSACEAE)**

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Most earth tongues are regarded as rare and endangered in most regions of their occurrence. Long-term research into these fungi, resulting in the delimitation and description of seven new species of *Glutinoglossum* in 2017, has continued in search of missing taxa. *Glutinoglossum glutinosum*, the genus type species, had not been recorded in Slovakia – all stored specimens labelled under this name represent different taxa. The species was discovered among specimens collected in the Slanské vrchy Mts. in 2020. It seems to be the only record in the country, but it has been collected occasionally in the Czech Republic. Variability, distribution and background

of its discovery are presented and discussed. It should be noted that while searching for *G. glutinosum*, four new taxa were distinguished and described, while several others are prepared for description.

The research was supported by project VEGA 2/0061/19.

TEN NEW FUNGAL SPECIES DESCRIBED AT THE BiMM RESEARCH PLATFORM (TULLN, AUSTRIA) DURING 2018–2021

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The research platform BiMM - Bioactive Microbial Metabolites was established in 2015 as a high-throughput approach to discover novel bioactive compounds by applying interactions between bacteria and filamentous fungi (co-cultivations). The microbial strains used in co-cultivations have been isolated by our team mostly from an environment (soil, water, air) or from clinical (veterinary and human) settings. During these microbial surveys, several new fungal taxa have been revealed and described based on the polyphasic approach. *Metapochonia lutea* (Ascomycota, Hypocreales) isolated from littoral water of the Danube river in Tulln (Austria) in 2017, was the first fungal taxon described by the BiMM team. *Saksenaea dorisiae* (Mucoromycota, Mucorales) was isolated from a water sample from a private well in Manastirica-Petrovac in the Republic of Serbia sampled in 2018. Four new *Keratinophyton* species (Ascomycota, Onygenales) were described this year, namely *K. gollerae* and *K. wagneri* isolated from forest soil (Tatranská Lomnica, SK, in 2019 and 2015, respectively), *K. lemmensii* from compost soil (Tulln, AT, in 2015), and *K. straussii* from garden soil (Vieste, I, in 2015). This year, also two new toxigenic members of *Penicillium* section *Exilicaulis* (Ascomycota, Eurotiales) were described, i.e. *P. krskae* isolated from air as a lab contaminant in Tulln (AT) and *P. silybi* isolated as an endophyte from asymptomatic milk thistle stems in Josephine County (Oregon, USA). In addition to these already published taxa, two putatively novel ascomycete species belonging to *Gymnoascella* (Onygenales) and *Flavocillium* (Hypocreales) found in Austria in 2021 are briefly described and illustrated herein.

Funding: The Bioactive Microbial Metabolites research platform (BiMM) is supported by grants K3-G-2/026-2013 and COMBIS/ LS16005 funded by the Lower Austria Science and Education Fund (NfB).

**SPECIES DELIMITATION IN *ASPERGILLUS*:
REVISION OF SECTION *FLAVIPEDES* AND MORE**

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The section *Flavipedes* of genus *Aspergillus* was revised in 2015. Since then, some taxonomic changes have been made in the section with four new species being described, and also a rapid development in the field of species delimitation has taken place. Because of this, we decided to carry out a new revision based on 90 strains belonging to section *Flavipedes*. We obtained sequences of three genes (beta-tubulin, calmodulin, and RPB2; the ITS sequences not being used because of limited variability) and utilised them for a phylogenetic analysis consisting of 2 parts. In the first part we used a four single-locus (GMYC, bGMYC, PTP, bPTP) and one multi-locus (STACEY) species delimitation method to devise the boundaries between hypothetical species. In the second part, we validated these hypothetical species with the recently developed DELINEATE programme instead of the traditionally used BPP. Aside from the phylogenetic analysis, we also performed classical macro- and micromorphological analyses testing the growth rate of selected strains at temperatures of 10 to 40 °C. As a result, we discovered and described four new species (*A. alboluteus*, *A. alboviridis*, *A. inusitatus* and *A. lanuginosus*) and synonymised *A. capensis* with *A. iizukae*. The revised section *Flavipedes* now harbours 18 species. Its representatives are mostly isolated from soil, but many strains in this study also originated from other substrates, such as indoor environment, clinical material, food, feed, and animal droppings.

TOMENTELLOID FUNGI OCCURRING IN SOUTH BOHEMIA

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Tomentelloid fungi, which include genera *Tomentella*, *Pseudotomentella*, *Tomentellopsis* and *Amaurodon*, are a group of the family Thelephoraceae creating resupinate fruitbodies. Although they had been considered saprotrophic for a long time, they were found to be ectomycorrhizal later and are supposed to be rather important in forest ecosystems as their ectomycorrhiza occur on roots of trees significantly. Despite their abundance they have been much overlooked because of the corticioid appearance of their fruitbodies and their relatively difficult identification. Therefore relevant data on the occurrence of tomentelloid species in the Czech Republic are missing. The aim of my work was to find out which tomentelloid species occur in South Bohemia and to determine their distribution, phenology and ecology, including potential preferences for substrates, habitats and ectomycorrhizal trees.

A total of 46 species of tomentelloid fungi have been identified so far, including 32 species of *Tomentella*, nine species of *Pseudotomentella*, four species of *Tomentellopsis*, and one species of *Amaurodon*. Five species are contained in the Red List of macromycetes in the Czech Republic, *Amaurodon atrocyaneus* (CR), *Pseudotomentella atrofusca* (?EX), *Pseudotomentella nigra* (CR), *Tomentella galzinii* (CR) and *Tomentellopsis zygoesmoides* (CR). Preferences of tomentelloid species for particular habitats and ectomycorrhizal trees have been confirmed, but substrate does not seem to be important for the occurrence of particular species. Differences in phenology between species and even genera of tomentelloid fungi are obvious as well.

SPECIMENS OF THE GENUS *HYPOMYCES* IN THE PRM HERBARIUM

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Some interesting specimens studied or identified during a revision of the genus *Hypomyces* in the herbarium of the National Museum are presented. The species

Sphaerostilbella berkeleyana and *S. broomeana*, found among unidentified specimens of the genus, are probably new to the Czech Republic. The former is represented by specimens collected by Dr. Svrček on *Stereum hirsutum* at Mezná near Hřensko and Dobřichovice, the latter by a specimen collected by Dr. Pouzar on *Heterobasidion parvisporum* in the vicinity of Stříbrná Skalice. In terms of taxonomy, the studied species *Hypomyces sepulchralis* and *Mycogone cervina* are worth mentioning.

GLOBALFUNGI – SVĚTOVÁ DATABÁZE ROZŠÍŘENÍ HUB

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Thanks to the recent advance in high-throughput-sequencing methods we are facing an accumulating wealth of fungal sequencing data from various geographical regions, ecosystems and habitats. Although the application of NGS methods has revolutionised our understanding of fungal ecology, the accumulating raw fungal NGS data in sequence repositories has not provided much extra value so far. The idea behind the GlobalFungi Database (<https://globalfungi.com/>) is to provide everyone access to published data on fungal community composition obtained by next-generation-sequencing through a web-based interface which allows various database queries and visualisation of the results. To date, we have collected more than 1 billion observations of fungal ITS1 and ITS2 marker sequences from next-generation-sequencing data published in 367 studies and containing more than 36,000 samples from around the world. Our database covers data from all terrestrial habitats except those subject to experimental manipulation, containing information on fungal communities from soil, litter, dead plant material, living plant tissues, water, air, dust, etc. GlobalFungi invites the scientific community to participate and encourage the entering of data by authors of studies which are not yet covered.

RIBOSOMAL DNA – BETTER THE DEVIL YOU KNOW

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DNA regions coding nuclear ribosomal RNA (rDNA) are the first-choice molecular markers with wide numerous applications in mycology. The lecture summarises the limits of rDNA usability for fungal identification and phylogeny inference. The ITS rDNA (Internal Transcribed Spacer region) can be newly used for the study of species geography and ecology by datamining of metabarcoding data using GlobalFungi database (<https://globalfungi.com/>). In this respect, there is a limit to ITS sequence variability, which often disables differentiation between distinct species which differ substantially in ecology and geographical distribution. Other risks lie in the little known issue of evolution speed (i.e. heterotachy) and GC content (i.e. guanine and cytosine content of DNA) heterogeneity across the datasets and even between related taxa. Both phenomena are affected by numerous neutral mechanisms, but also by evolution transitions to new life styles and changes in population sizes. Heterotachy and GC content bias, affecting the whole genome, also dramatically affect rDNA applicability to phylogenetic studies. These biases are demonstrated on the case of *Kurtia* (formerly *Hyphoderma*, Hymenochaetales, Basidiomycota) and some lineages of rusts and smuts.

PRACTICAL (AND TRAGIC) CONSEQUENCES OF IDENTIFICATION OF FUNGI BASED ON DNA BARCODES ONLYOndřej K o u k o l ¹, Greg D e l g a d o ²

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The knowledge of (1) recent and historical literature, (2) diagnostic characteristics including their variability, and (3) ecology including distribution and host or

habitat specificity represents key skills in fungal taxonomy and above all in studies dealing with microscopic fungi. At present, these traditional pillars are combined with molecular data analyses. Growing availability and decreasing costs of obtaining this data are, however, leading to a predominant if not the exclusive use of sequence similarities in fungal identification as well as the delimitation of new species or higher taxonomic categories. In our contribution, we present several examples from recent history when obviously erroneous taxonomic conclusions were drawn because too much attention was paid to the analysis of molecular data. Morphological characteristics were only used as support for results based on molecular data without even mentioning key literature sources. Despite being based on erroneous conclusions, these taxa were validly described and have now become obstacles for further taxonomical but also biodiversity studies. Correcting these errors is a difficult and time-consuming process.

CADMIUM HYPERACCUMULATION IN *THELEPHORA PENICILLATA* AND ITS ISOTOPIC COMPOSITION

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The highest concentrations of cadmium in mushrooms, reaching hundreds of milligrammes per kg of dry mass, have been reported in the Agaricaceae family, namely in *Agaricus crocodilinus* and *Cystoderma carcharias*. However recently, an astonishingly strong ability to accumulate toxic elements has been detected in *Thelephora penicillata* (Thelephoraceae). We found Cd concentrations in the range of 580 to 2060 mg·kg⁻¹, all samples originating from unpolluted sites. Moreover, *Thelephora penicillata* accumulates As (430–1470 mg·kg⁻¹) and also reduced increased levels of Cu and Zn (hundreds of mg·kg⁻¹).

In the vicinity of Kašperské Hory (West Bohemia) we investigated the isotopic composition of Cd [$\delta^{114/110}\text{Cd}$ (‰)] in *Thelephora penicillata* and associated soil samples, which we compared to that of other mushrooms growing at the same site.

The isotopic composition of *Thelephora penicillata* ($\delta^{114/110}\text{Cd}$ 0.030–0.140) corresponded roughly to that of soil 1M HNO₃ extracts (unmilled <2 mm fraction), which we believe is a fraction hypothetically accessible to the mushroom. Other ectomycorrhizal mushrooms growing at the same site (e.g. *Amanita* spp., *Cantharellus cibarius*, *Boletus* s.l.) were often characterised by distinct isotopic values, mostly isotopically lighter (–0.374 to 0.139), which indicates mass-dependent isotopic fractionation of Cd during the accumulation process (Cd mobilisation in soil by bioleaching – mycelial uptake – mycelial transport – accumulation in fruitbodies).

Preliminary results indicate that possibly only a minor amount of Cd in *Thelephora penicillata* is sequestered by metallothioneins (MT) or MT-like peptides; a major fraction of intracellular Cd is likely sequestered in vacuoles or similar structures, probably in a complex with glutathione. Both inorganic As and methylated organoarsenicals were detected in the fruitbodies.

This research was supported by the Czech Science Foundation (project 19-06759S).

APPLICATION OF FLOW CYTOMETRY IN THE STUDY OF ECOLOGY AND FUNCTIONAL TRAITS OF LICHENS

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The nuclear genome size of eukaryotic organisms differs by a factor of more than 80,000. However, it was soon discovered that genome size is not correlated with the complexity of an organism, as an increase in genome size is often associated with an expansion of non-coding repetitive DNA, which may occupy more than half of the genome. At first, it was assumed that organisms with a high proportion of repetitive sequences in the genome simply lack efficient defence mechanisms against their spread. However, it was soon revealed that the development of an organism is not only affected by its genes, but also by its genome size as a whole, as genome size affects

a number of functional traits such as cell size, cell division and metabolic rate. Thus, genome size is now considered an important trait in species adaptations to environmental conditions. So far, we only have sparse information on fungal genome sizes and their impact on fungal ecology. Our goal is to provide a first insight into lichen genome sizes using the flow cytometry approach and to study changes in genome sizes in relation to their ecology (e.g. preferred substrate) and functional traits (e.g. thallus type, reproduction strategy, fruitbody and spore size). So far, we have measured genome sizes of 150 lichen species of several dozen families representing the Central European lichen flora. Preliminary data suggests a positive correlation between genome size and spore size. Furthermore, foliose lichens have larger genomes than fruticose and crustose ones. The most interesting finding is the difference between r- and K-strategists, where r-strategists have smaller genomes than K-strategists. The small genomes of r-strategists may promote rapid growth, which is advantageous in the colonisation of ephemeral substrates.

**ASCOSPORE MORPHOLOGY OF BRYOPHILOUS PEZIZALES
IS CLOSELY ASSOCIATED WITH HOST ECOLOGY
AND POSITION OF INFECTION**

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Since fungal spores exhibit a high diversity in size, shape, number of cells and ornamentation, they are frequently used as diagnostic traits in taxonomy. However, only little is currently known about the possible function of spore traits and the relationship between spore morphology and fungal ecology. Bryophilous Pezizales, biotrophic parasites of bryophytes, represent a suitable model system for the study of this question, because their species differ significantly in ecology and spore morphology.

We analysed 130 different representatives of this fungal group and investigated the relationship between their ascospore traits, host ecology and dominant position of infection using the phylogenetic generalised least squares technique. Species attached to the rhizoids of bryophytes had ascospores with a significantly more globose shape, higher ornamentation, and higher relative content of lipid bodies than species physically connected to leaves and stems. In addition, higher ornamentation and higher relative content of lipid bodies also occurred in species connected to bryophytes with a shorter lifespan.

Several of the detected relationships could reflect different requirements for persistence or spore dormancy and specific soil conditions affecting species connected to rhizoids. Our results thus suggest that the studied ascospore traits play an important role in the ecology of this fungal group and may help us understand also better the importance of spore morphology in other fungi.

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DETECTION OF MICROMYCETES ON BERRIES FROM SLOVAK SUPERMARKETS

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In the year 2021, we monitored the occurrence of micromycetes in samples (29) of fresh berries obtained from Slovak supermarkets. A total of 66 isolates of micromycetes belonging to 7 genera were isolated. Samples of strawberries (7) contained representatives of the genera *Botrytis* (isolation frequency (Fr) 85.7%, relative density (RD) 46.2%), *Rhizopus* (Fr 28.6%, RD 15.4%), *Cladosporium* (Fr 14.3%, RD 7.7%) and *Penicillium* (Fr 57.1%, RD 30.8%: *P. hordei*, *P. fellutanum*, *P. atramentosum* and *P. expansum*). Samples of raspberries (12) contained representatives of the genera *Botrytis* (Fr 58.3%, RD 20%), *Cladosporium* (Fr 100.0%, RD 42.9%), *Rhizopus* (Fr 8.3%, RD 2.9%), *Mucor* (Fr 8.3%, RD 2.9%) and

Penicillium (Fr 66.7%, RD 31.4%: *P. citrinum*, *P. pulvis*, *P. digitatum*, *P. brevicompactum*, *P. kiamense*, *P. purpurescens*, *P. bialowiezense* and *P. expansum*). Samples of blackberries (5) contained representatives of the genera *Cladosporium* (Fr 40.0%, RD 50.0%), *Botrytis* (Fr 20.0%, RD 25.0%), *Penicillium* (Fr 20.0%, RD 25.0%: *P. brevicompactum*) and blueberry samples (5) included representatives of the genera *Botrytis* (Fr 100.0%, RD 35.7%), *Cladosporium* (Fr 80.0%, RD 50.0%), *Alternaria* (Fr 20.0%, RD 7.1%) and *Aspergillus* (Fr 20.0%, RD 7.1%). Four isolates of potentially production strains of the genus *Penicillium* associated with fruit degradation were tested with thin layer chromatography for their ability to produce selected mycotoxins. All tested isolates of *P. citrinum* (2) produced citrinin and all tested isolates of *P. expansum* (2) produced patulin, citrinin, roquefortin C and cyclopiazonic acid.

The contribution was financially supported by projects VEGA/0517/21 and KEGA 022SPU-4/2021.

ANTAGONISTIC EFFECT OF ENDOPHYTIC FUNGI FROM EUROPEAN ASH TREES AGAINST *HYMENOSCYPHUS FRAXINEUS*

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Ash dieback caused by the fungus *Hymenoscyphus fraxineus* seriously endangers forest stands in Slovakia. There are very limited ways of effective control of ash dieback in forests. In the present study, we assessed the inhibitory effect of endophytic fungi naturally occurring in ash trees against *H. fraxineus* in the form of dual in vitro cultures. Endophyte isolates were obtained by incubating segments of asymptomatic leaves and branches on malt extract agar. The samples were collected in a European ash forest close to the town of Nitra (SW Slovakia) in May and October 2019. A total of 76 isolates of endophytic fungi colonising leaves (31 isolates) or branches (45 isolates) were obtained, which we identified by means of sequence analysis of the ITS region of rDNA. We identified 26 species of 21 genera, dominated by species of the genera *Alternaria* (25 isolates), *Diaporthe* (10), *Phoma* (10), *Epicoccum* (4) and *Fusarium* (4). The dual culture assay was carried out in Petri dishes

(90 mm) on malt extract agar supplemented with extract of ash leaves (for leaf endophytes) or sawdust of ash branches (for branch endophytes) for 27 days at 25 °C. The diameter of *H. fraxineus* colonies was measured at the end of the experiment and compared with the diameter in the control with dishes of *H. fraxineus* monoculture. The assay was carried out in three replicates and the effect of endophytes on *H. fraxineus* colonies was evaluated using analysis of variance. Four endophyte isolates from branches (*Fusarium lateritium*, *Phoma aliena*, *Dothiorella gregaria*, *Diaporthe macrostoma*) and one endophyte from leaves (*Alternaria alternata*) significantly ($p < 0.05$) inhibited the growth of *H. fraxineus*. The growth of *H. fraxineus* colonies was reduced by 33–57%.

The work was supported by grant agency VEGA, project no. 2/0062/18.

MYCOTOXINS AND BEER

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Mycotoxins are secondary metabolites of moulds, representing a health risk to humans. A good check of raw materials in beer production is as important as the control of the final product because mycotoxins can be transferred from contaminated barley to the malt and subsequently to the beer. The occurrence of deoxynivalenol and ochratoxin A was monitored in commercial brands of beer in the years 2017–2019. Deoxynivalenol was analysed by means of liquid chromatography with mass spectrometry (LC-MS/MS) and ochratoxin A by means of liquid chromatography with fluorescent detection (UPLC-FLR). Both deoxynivalenol and ochratoxin A were found in 85% of samples ranging from 1.01 to 25.20 µg/l and 1.4 to 95.6 ng/l, respectively.

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**ISOLATION OF *DALDINIA* SPECIES
FROM TRUNK TISSUES OF *BETULA PENDULA***

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In the year 2020 in the tree nursery of Arboeko (Smržice, Czech Republic) symptoms of suddenly decaying 5-year old birch trees planted in optimal soil and climate conditions were observed. These included 5–10 centimetre long lesions on the surface of the trunks and 1.5 centimetre wide spotted necrosis on the cross-section of the trunk.

From the lesions of two trees 40 wood samples were collected for traditional cultivation methods on PDA and MEA media, from which 21 fungal isolates were cultivated. These fungal isolates were identified according to culture morphology and sequencing of the ITS region of the ribosomal RNA gene, LSU (large subunit rDNA) and protein-coding genes RPB2 (encoding the second largest subunit of RNA polymerase II) and TUB2 (β -tubulin) as proposed by Wongkanoun et al. (2020).

A pathogenicity test was performed on branches (15–20 cm) taken from healthy birch trees.

Pathogenic behaviour of *Daldinia* species has been described in wood tissues of *Betula pendula* L. and other deciduous trees such as beech (*Fagus sylvatica* L.), oaks (*Quercus* spp.), ash (*Fraxinus excelsior* L.) and maple (*Acer platanoides* L.).

The observed lesions on the trunk of birch trees were probably caused by the detected fungus *Daldinia decipiens*.

**TWO NEW MEADOW SPECIES OF THE FAMILY HYGROPHORACEAE
FOUND IN SLOVAKIA**

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European grassland species of the family Hygrophoraceae are well known as bioindicators. So far more than 60 species have been described from Europe. We present two newly described species – *Hygrocybe fulgens* and *Neohygrocybe* sp. nov., which were found at several localities in Slovakia. *Hygrocybe fulgens* is characterised by a dry, squamulose pileus, medium-sized basidiomata, early occurrence (July to September), ellipsoid spores and orange-red basidiomata (sometimes with golden yellow hues). *Neohygrocybe* sp. nov. is characterised by pale brownish grey basidiomata with a nitrous smell, its flesh not changing colour, a compressed and hollow stipe, sometimes filled with a cottony substance.

The research was funded by the Operational Programme Integrated Infrastructure, co-financed by the European Fund for Regional Development under project (EFRD) ITMS2014+313021W683: “DNA barcoding of Slovakia (SK-BOL), as a part of international initiative International Barcode of Life (iBOL)” and by the Grant Agency for Research and Development under project no. APVV-17-0317.

FUNGAL SECONDARY METABOLITES IN MICROBIAL COMMUNITY INTERACTIONS

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Most herbivorous insects are unable to digest much of the plant biomass, which in addition contains a number of antiherbivore substances (tannins, flavonoids). Biodegradation of plant biomass in the intestines is therefore mediated by a diverse community of intestinal microbiota. One of the characteristics of complex communities like intestinal microbiota are interactions between partners mediated by secondary metabolites of microorganisms. In our experiments, a selected set of dominant fungi of the intestinal microbiota of caterpillars was individually cultivated in media with antiherbivore substances. The biological activity of crude extracts of the media and changes in biomass weight and metabolic activity were then determined. Some of the species of the genera *Aureobasidium*, *Aspergillus* and

Exobasidium reacted to these substances by increasing their metabolic activity (higher biomass, induction of secondary metabolite production). As an example, the production of new antimicrobial guanin derivatives were triggered in *Exobasidium* sp. by adding antiherbivore substance (rutin). The study could provide us important information on plant-animal-microbial interactions but the results will be also important from the perspective of biotechnology.

DETECTION OF MYCOVIRUSES IN SLOVAK *HYMENOSCYPHUS FRAXINEUS* POPULATIONS

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The massive mortality of ash trees caused by *Hymenoscyphus fraxineus* endangers the existence of *Fraxinus* species on the European continent including Slovakia. At present, biological and environment-friendly methods gain ground in plant protection. Mycoviruses (ssRNA or dsRNA) used in biological control decrease the virulence of phytopathogenic fungi such as *Cryphonectria hypovirus 1* (CHV-1) in chestnut blight disease. We assume that the fungus *H. fraxineus* could be infected by mycoviruses, which might also have influence on the virulence of the fungus and could consequently be used for biological control of ash dieback.

The aim of our study was to determine the presence of mycoviruses in Slovak *H. fraxineus* populations. In total, 45 isolates of *H. fraxineus* obtained from lesions of common ash (*Fraxinus excelsior*) from 12 localities in Slovakia were analysed. Mycoviruses were detected using dsRNA isolation. The presence of dsRNA was confirmed in eight isolates. In three isolates the dsRNA band of 2.5 kb was observed, whereas the dsRNA band of 2.2 kb was observed more frequently, in five isolates. These isolates were tested for the presence of *Hymenoscyphus fraxineus mitovirus 1* (HfMV1) with RT-PCR using specific primers Cf4_F1 and Cf4_R3. The PCR

products were ca 500 bp in length. These results confirm the presence of HfMV1 in the examined isolates, which we plan to test for biological protection of ash.

This research was funded by the Scientific Grant Agency of the Slovak Ministry of Education and by Slovak Academy of Sciences grant VEGA 2/0062/18.

ANTIMONY AND ARSENIC IN MACROMYCETES AT LOCALITIES AFFECTED BY ANTIMONY MINING IN SLOVAKIA

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In 2018 and 2019, macromycete fruitbodies and soil samples were collected at three localities affected by antimony mining: Dúbrava and Medzibrod in the Nízke Tatry Mts., and Čučma in the Spišsko-Gemerské Rudohorie Mts. The concentration of potentially toxic elements in the fruitbodies was determined with the ICP-MS method and in the soil with the ICP-MS and ICP-ES methods. The bioconcentration factor (BCF) was determined in selected samples. The highest concentration of arsenic was found in the fruitbodies of *Cortinarius glaucopus* (329.2 mg·kg⁻¹, BCF 2.24) from the Dúbrava locality. Extremely high values were also measured in *Paxillus olivellus* (169.6 mg·kg⁻¹) and *P. rubicundulus* (167.9 mg·kg⁻¹) from Medzibrod. Of the edible species, the arsenic content greatly exceeded the permitted limit in *Gomphidium glutinosus* (34.0 mg·kg⁻¹) and *Lactarius deterrimus* (16.2 mg·kg⁻¹) from Dúbrava. The fruitbodies of the popular edible species *Suillus grevillei* contained the highest concentrations of antimony (363.33 mg·kg⁻¹), but high concentrations were found in all samples of different species of the genus *Suillus* from Dúbrava (*S. grevillei*, *S. luteus*, *S. granulatus*). The highest concentrations of antimony were found in the species *Amanita gemmata* (49.23 mg·kg⁻¹), *Paxillus rubicundulus* (109.76 mg·kg⁻¹) and *P. olivellus* (82.06 mg·kg⁻¹) at Medzibrod and in *Echinoderma asperum* (83.78 mg·kg⁻¹) at Čučma. BCF was >1 in three samples of *Suillus* from Dúbrava. Collection of edible mushrooms for consumption at the monitored localities is not recommended.

The research was funded by the Operational Programme Integrated Infrastructure, co-financed by the European Fund for Regional Development under project (EFRD) ITMS2014+313021W683 “DNA barcoding of Slovakia (SK-BOL), as a part of international initiative International Barcode of Life (iBOL)” and by the Grant Agency for Research and Development under project no. APVV-17-0317.

EX-TYPE CULTURES IN THE CULTURE COLLECTION OF FUNGI (CCF)

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The Culture Collection of Fungi (CCF) at the Department of Botany, Faculty of Science, Charles University Prague preserves microscopic saprotrophic fungi. It focuses mainly on ascomycetes and zygomycetes and serve as documentation and comparative material for the taxonomic work of researchers and students of the Department of Botany. It also provides strains to other education and research institutions. Ex-type cultures (i.e. from type-derived cultures) belong to the most valuable parts of culture collections. The total number of strains in the CCF is now 4,884 (data until 30 June 2021), of which 452 belong to ex-type cultures. Most of these ex-type strains were obtained from abroad (328, i.e. 73%) but a significant part (124, i.e. 27%) originates from the research activities of staff and students of the Department of Botany, Charles University. These ex-type strains are stored in the CCF in liquid nitrogen and in a lyophilised state.

Most ex-type cultures belong to the genera *Penicillium* (161) and *Aspergillus* (143). *Penicillium camemberti* and *P. commune* (both isolated in 1904), *P. atramentosum* and *P. rolsfii* (isolated in 1905), and *Talaromyces rugulosus* (isolated in 1906) belong to the oldest fungi stored in culture collections. Thirty-eight ex-type cultures come from the Czech Republic.

The work was financially supported by the Ministry of Agriculture of the Czech Republic (The National Programme on Conservation and Utilization of Microbial Genetic Resources and Invertebrates of Agricultural Importance) and the Czech Ministry of Education, Youth and Sports (Institutional support).

**IMPACT OF FOREST STRUCTURE
ON THE DIVERSITY OF WOOD-INHABITING FUNGI
IN PRIMARY FORESTS OF THE CARPATHIAN MOUNTAINS**

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Wood-inhabiting fungi are lignicolous organisms associated with deadwood, which they use as a source of nutrition or a substrate for growth. In forest ecosystems, they play an important role in the decomposition of wood and participate in the nutrient cycle. They are endangered mainly by habitat destruction and the removal of deadwood. Fairly little is known about their ecology and distribution, and research is mainly focused on managed forests. In order to prevent further loss of biodiversity, it is necessary to know more about the functioning of communities in natural conditions, where the structure of the stand is the result of long-term effects of natural forest dynamics.

For this purpose, 117 permanent circular plots with a radius of 17.84 m were established in spruce and mixed beech forests in Slovakia. Based on the presence of fruitbodies, all macromycetes were recorded on selected deadwood objects during one visit to each plot in autumn 2017 (spruce forests) and 2019 (mixed beech forests). Structural and dendrochronological characteristics were also recorded for all research areas under the REMOTE project.

Approximately 650 fungal taxa were recorded. The number of species in beech forests was more than twice as high as in spruce forests. Many rare species were found (e.g. *Amylocystis lapponica*, *Fomitopsis rosea*, *Auriporia aurulenta*, *Hydropus atramentosus*, *Skeletocutis odora*). We will further evaluate the investigated diversity of

fungi in relation to the structure and history of forest stands using dendrochronological methods. The results can help make the protection of the remaining forests more effective, but they can also be used as a basis for close-to-nature forest management.

SPECTRUM OF FIELD MICROMYCETES ON WHEAT OF SLOVAK ORIGIN

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The study dealt with monitoring the occurrence of microscopic filamentous fungi on wheat grains of Slovak origin, grown in 2019 and 2020 at various localities of the Nitra, Banská Bystrica and Prešov regions. The main interest was to analyse the spectrum of potential mycotoxin producers in field conditions, especially representatives of the *Alternaria* and *Fusarium* genera. A total of 15 wheat samples were analysed by direct placing of superficially sterilised grains on agar plates with dichlorane, Rose Bengal and chloramphenicol (DRBC). The *Alternaria* genus was detected at an isolation frequency (Fr) of 100% and an average relative density (RD) of 46.4%. Overall, the genus represented the highest proportion of isolated fungi. RD in individual samples ranged from 4.3 to 76.3%. Four species groups were identified: *A. alternata*, *A. arborescens*, *A. infectoria* and *A. tenuissima*. The *Alternaria tenuissima* species group was the most common representative of this genus (Fr 100%, average RD in the genus 59.8%). The *Fusarium* genus was isolated from wheat samples at an Fr of 87% and an average RD of 5.0%. The RD of this genus in individual samples was in the range of 0.0–24.4%. A total of 11 species were isolated: *F. avenaceum*, *F. culmorum*, *F. equiseti*, *F. fujikuroi*, *F. graminearum*, *F. incarnatum*, *F. langsethiae*, *F. oxysporum*, *F. poae*, *F. proliferatum* and *F. sporotrichioides*. *Fusarium avenaceum* was isolated at the highest Fr (40%). In the observed genus, *F. avenaceum* (27.7%), *F. graminearum* (22.9%) and *F. proliferatum* (20, 5%) species were detected at the highest average RD.

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MYCOCENOSIS OF SELECTED HISTORICAL MONUMENTS IN BRATISLAVA

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Changing climatic conditions pose new challenges to spatial planning. Standard measures of urban space management need to be expanded with new knowledge based on analyses of objects exposed to extreme weather conditions. Historical monuments have a specific position in settlements, as legislation limits the application of protection measures. In this research, we observed two historical monuments located in the centre of Bratislava: the charnel-house of Saint James's Chapel (13th century) and a Celtic kiln dating from the 1st century BC. Both buildings are situated below the ground and are partially protected by glass coverings (a shelter and a wall). Ventilation of the premises and maintenance of a stable humidity level are not sufficiently secured, and atmospheric precipitation has penetrated the buildings.

We present the results of an initial mycological analysis. We monitored the occurrence of species and simultaneously major climatic indicators (temperature, humidity).

From the surface of the skulls in Saint James's Chapel, we identified the species *Acremonium* sp., *Aspergillus niger*, *A. ochraceus*, *A. clavatus*, *Cladosporium cladosporioides*, *Cladosporium* sp., *Monocillium mucidum*, *Paecilomyces* sp., *Penicillium* sp., *Pithomyces chartarum*, *Trichoderma* sp. and sterile light-coloured mycelium.

From the surface of the Celtic kiln we isolated the species *Aspergillus ochraceus*, *A. oryzae*, *Cladosporium cladosporioides*, *Gilmaniella humicola*, *Penicillium* sp., *Scopulariopsis asperula*, *Trichoderma* sp. and *Rhizopus stolonifer* var. *stolonifer*.

Both objects had a sufficient source of organic substances for the occurrence of microscopic fibrous fungi. The localities are open to visitors, so micro-organisms get into the premises of historical buildings by the flow of air and the clothes of visitors. The identified species are also known from other monuments. They cause bio-deterioration and subsequent biodegradation. Coupled with precipitation and extreme

temperatures (especially in summer during heat waves), this damage the quality of historical buildings.

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ENDOPHYTIC MYCOBIOTA OF VINE ROOTS – FIRST RESULTS

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In the spring of 2021, a study into the endophytic mycobiota of roots of grapevines grown in the Czech Republic was initiated. So far, 195 roots of 30- to 40-year-old shrubs from 4 locations (two in Central Bohemia and two in South Moravia) have been investigated. The obtained fungal strains have been identified on the basis of micro- and macromorphology on genus or morphotype level. Identification to species level using sequencing of selected sections is in progress. Fungi belonging to the former genus *Cylindrocarpon* found in 32.36% of the segments were the most frequent at all sites. *Clonostachys rosea*, species from genera *Fusarium*, *Phomopsis* and *Cladosporium* and more than 25 morphotypes of fungi (in the form of sterile mycelia) were detected at significantly lower frequency.

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TESTING OF INHIBITORY EFFECTS OF FUNGICIDES AS PREVENTION AGAINST ASH DIEBACK CAUSED BY *HYMENOSCYPHUS FRAXINEUS*

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According to the biology of *Hymenoscyphus fraxineus* (causal agent of ash dieback), trunk injection seems to be a suitable alternative to chemical control method. The inhibitory effect of three fungicides with dodine (Syllit 65 WP: 1.91 g/l at a dose of 2 ml per cm of DBH – diameter at breast height), sulphur (Kumulus WG: 4.3 g/l at a dose of 2 ml/cm of DBH), and potassium phosphate (Enerbite: 1 g/cm of DBH) as the active compounds was evaluated. Application of these fungicides to the stems of 15 year old ash trees (*Fraxinus excelsior*) was realised using the BITE® (Blade for Infusion in TrEes) system for 12 weeks before inoculation of *H. fraxineus* to host trees. The inhibitory effect was recorded only on selected trees injected with sulphur (Kumulus WG) and potassium phosphate (Enerbite). The average area of necrotic lesions on these trees reached 6.25 cm² and 6.16 cm², respectively, in comparison with 13.06 cm² on the control. The differences were not statistically significant due to the high variability of necrotic wound areas in the compared groups. No inhibitory effect was observed for dodine (Syllit 65 WP).

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MICROMYCETES COLONISING WINTER WHEAT STEMS IN SLOVAKIA

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Cereals are important crops grown for grain in Slovakia. Their production is significantly influenced by the activity of parasitic microscopic fungi under the agroecological conditions of the country. The aim of this study was to identify the mycobiota of winter wheat (*Triticum aestivum*) stems based on the morphological characteristics of fungi detected on plant material in the stage of full physiological maturity of the grain. Wheat plant samples were collected at 162 localities in wheat production areas of Slovakia during the years 2018–2020. The results of mycological analysis include characteristics of the species diversity of micromycetes and their frequency of occurrence in the examined samples. Members of ascomycetes (58%)

and deuteromycetes (42%) were identified in the samples. The most frequently identified fungi were the following 12 species (frequency of occurrence in brackets): *Pyrenophora tritici-repentis* (72.8%), *Gibberella zeae* (66.7%), *Monographella nivalis* (57.4%), *Lewia infectoria* (42.0%), *Phoma* sp. (36.4%), *Fusarium* sp. (32.7%), *Colletotrichum graminicola* (32.1%), *Lophodermium gramineum* (30.2%), *Phaeosphaeria herpotrichoides* (26.5%), *Blumeria graminis* (25.9%), *Gaeumannomyces graminis* (22.8%), *Bipolaris sorokiniana* (15.4%).

The activity of parasitic fungi infesting particular parts of wheat plants manifested itself by the formation of necrotic lesions, rot and dying of stems, and ultimately by the lodging of winter wheat stands. The species *Monographella nivalis*, *Gibberella zeae* and *Gaeumannomyces graminis* were identified as causal agents of rot and dying of the winter wheat stem base in Slovakia.

This work is an output from Research and Development (R&D) project Characterization of Plant Genotypes and Interacting Communities of Microorganisms in Changing Climatic Conditions funded by the Ministry of Agriculture and Rural Development of the Slovak Republic.

**CRYSTALS AS AN OVERLOOKED CHARACTER IN THE GENUS
PSEUDOPLECTANIA, AND A NEW SPECIES OF *PSEUDOPLECTANIA*
FROM SOUTH AFRICA**

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An undescribed species of *Pseudoplectania* (Sarcosomataceae) was found on a decaying coniferous trunk during an excursion in South Africa. Microscopically, it was striking due to the presence of large yellow crystals in the hymenium and ectal excipulum, so far known only in *P. tasmanica*. The aim of our work was to describe

the new species officially, compare it with other species of the genus and evaluate the presence of crystals in available species of *Pseudoplectania*.

The name *Pseudoplectania africana* Carbone & Sochorová has been suggested for the species. Its characteristic features are sessile or shortly stipitate apothecia which are brown to black, sometimes with an olive tinge, up to 18 mm broad, and have rich basal tomentum. Microscopical features are – besides the crystals – smooth ascospores with an eccentric, relatively small sheath, straight, often bifurcate paraphyses, and wavy to coiled hairs. The same type of crystals as in *P. africana* and *P. tasmanica* was newly found in *P. ericae*. These three species are closely related. The crystals in the hymenium are very abundant, angular, cracked, sometimes very long (often over the whole layer of the hymenium). They are not soluble in any of the tested media (Melzer's solution, 40% KOH, 9% HCl, 60% ethanol). In UV light, they are sulphur yellow. The crystals in the ectal excipulum are very similar, only smaller. The other tested species (*P. nigrella*, *P. lignicola*, *P. melaena* and *P. episphagnum*) only contain small hyaline crystals. In UV light, the whole section is coloured in shades of blue. *Pseudoplectania africana* is the first published record of *Pseudoplectania* from South Africa, and probably also from the whole of continental Africa.

Furthermore, type material of *P. episphagnum* was successfully sequenced during this study and the first sequences of *P. lignicola* were obtained.

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**CORRELATION BETWEEN MULTILOCUS GENOTYPE, CLINICAL
PICTURE AND MORPHOLOGY OF *TRICHOPHYTON MENTAGROPHYTES* /
T. INTERDIGITALE ISOLATES**

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Trichophyton interdigitale is one of the most common agents of tinea pedis and onychomycosis in humans. The closely related species *T. mentagrophytes* is the prime agent of superficial infections in animals (e.g. rabbits, cats and dogs), but it also commonly causes zoonotic infections in humans, especially tinea corporis. Based on the original concept, these species should be distinguishable by typical phenotypic features, including macromorphology of colonies, presence/absence of macroconidia and spiral hyphae, etc. However, recent studies have indicated that the correlation between the clinical picture and phenotype and genotype of the pathogen is not so clear as had been expected. Due this, the differentiation of these taxa mostly relies on several substitutions in the internal transcribed spacer region.

A total of 120 isolates obtained from Czech patients with various clinical manifestations of dermatophytosis were studied. A correlation analysis of morphology, clinical picture and molecular characterisation of three loci, ITS, β -tubulin and translation elongation factor 1- α (TEF), was performed.

Correlation was found between genotypic lineage *T. interdigitale* and factors such as clinical manifestation of tinea pedis / onychomycosis, age of patient and growth rate at 37 °C.

Mating-type genes were also characterised. The genotypic lineage *T. interdigitale* consisted exclusively of MAT-1-2 (HMG), whereas genotypic lineage *T. mentagrophytes* consisted of either one of HMG mating types or an alpha-box gene. A recombination analysis found gene flow between the genetic lineages. The results of several species delimitation methods were compared.

INFLUENCE OF SELECTED ESSENTIAL OILS OF PLANTS OF THE FAMILY MYRTACEAE ON THE GROWTH OF STRAINS OF THE GENUS *RHIZOPUS*

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The aim of the research was to test the effect of essential oils (EOs) (cloves, eucalyptus, tea tree, kajeput and niaouli) prepared from plants of the family Myrtaceae on the growth of *Rhizopus stolonifer* (2 strains) and *Rhizopus lyococcus* (1) strains isolated from mouldy fruits and vegetables. The gas diffusion method was used to test

the antifungal activity of EOs. A spore suspension (10^6 spores in 1 ml) was inoculated into the centre of the Petri dishes. Sterile filter paper and $625 \mu\text{l}\cdot\text{l}^{-1}$ EO (oil volume / air volume) were placed in the lid of a Petri dish. Dimethyl sulfoxide (DMSO) was applied to the control dishes. Inoculated Petri dishes were sealed with a parafilm and cultivated at $25 \pm 1 \text{ }^\circ\text{C}$ for 7 days in the dark. Colony growth was measured after 2, 3, 4 and 7 days of cultivation. EOs which inhibited growth of the strain completely were used to determine the minimum inhibitory doses (MIDs). EOs dissolved in DMSO were then prepared at different concentrations (625, 500, 250, 125, 62.5, 31.25 and $15.63 \mu\text{l}\cdot\text{l}^{-1}$). MID was expressed as the lowest concentration of EO at which no visible growth of strains was observed after 7 days of cultivation (at $25 \pm 1 \text{ }^\circ\text{C}$ in the dark). All tested EOs affected the growth of the strains negatively. Only clove EO inhibited the growth of all tested strains. The strains responded differently to the presence of EOs. The growth of *R. lycococcus* KMi-512 was completely inhibited by four (clove, tea, kajeput and niaouli) EOs, but the MIDs were $625 \mu\text{l}\cdot\text{l}^{-1}$. The growth of *R. stolonifer* KMi-524 was completely inhibited by both clove EO (MID $500 \mu\text{l}\cdot\text{l}^{-1}$) and tea EO (MID $625 \mu\text{l}\cdot\text{l}^{-1}$). The growth of *R. stolonifer* KMi-510 was completely inhibited by both clove EO (MID $250 \mu\text{l}\cdot\text{l}^{-1}$) and eucalyptus EO (MID $625 \mu\text{l}\cdot\text{l}^{-1}$).

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CROSSING THE ATLANTIC: RESURRECTION OF *ALBATRELLUS SIMILIS*?

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There are many examples of erroneous applications of North-American species concepts to European samples within the polypores. The Atlantic Ocean represents a barrier only rarely overcome by fungi, which supports allopatric speciation. In 1966, Zdeněk Pouzar described the new species *Albatrellus similis* based on material from South Bohemia and then, in 1972, synonymised this species with *Albatrellus subrubescens* described in 1940 from Florida (as *Scutigera subrubescens*). Pouzar originally based his concept on Lowe's erroneous interpretation of *Scutigera subrubescens* as a synonym of *Albatrellus confluens*. Therefore only careful examination

of the type material of *Scutigera subrubescens* confirmed conspecificity with *Albatrellus similis*.

The revelation of relationships within *Albatrellus* sensu stricto based on molecular data is the aim of the presented poster. We emphasised a comparison of North-American and European samples now classified as *Albatrellus subrubescens*. Preliminary results partly confirm the assumption that both populations are phylogenetically separated, although the lineage with European specimens also includes several North-American specimens. This may indicate the possible role of humans crossing the barrier, in particular from Europe to North America. The obtained molecular data also showed a considerable rate of undescribed diversity in the genus *Albatrellus* sensu stricto.