

Extended studies on the diversity of arthropod-pathogenic fungi in Austria and Poland

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Results of studies on diversity of arthropod-pathogenic fungi in selected habitats in Austria and Poland carried out in the years 2006–2007 and 2009–2010 are discussed. In total 47 species of entomopathogenic fungi were found as pathogens of different arthropods in Austria. Twenty six entomophthoralean species from different insects and one species from mites were identified and 16 of them are recorded as new to Austria. From among 21 species of anamorphic Hypocreales (Ascomycota) affecting arthropods in Austria, 13 species so far have not been known from this country. In total 51 species of fungi affecting different arthropods in Poland were recorded, among them 28 species of Entomophthorales and 23 anamorphic Hypocreales were separated. The most frequent species of the entomopathogenic fungi both in agricultural and afforested areas in Austria were the common and usually worldwide distributed cordycipitaceous anamorphs *Beauveria bassiana*, *Isaria fumosorosea* and in areas of this study less numerous *I. farinosa*. The most frequent pathogens occurring in mite communities on plants and in wood infested by insects were *Hirsutella* species. Several entomophthoralean species developed epizootics that caused high reduction in host populations of different arthropods in both countries. Especially interesting is the first record of mycoses (up to 60% mortality), caused by *Zoophthora* spp. on *Phyllobius* beetles in a mixed forest near Białowieża. During our joint research, we found the first time in Poland and Europe, the presence of the fungus *Furia* cf. *shandongensis* on earwigs and *Hirsutella entomophila* on *Ips typographus* adults in forest habitats. From the feeding sites of the latter bark beetle and other subcortical species in oak bark (mostly *Dryocoetes villosus*) and *D. alni* in black alder over a dozen of various *Lecanicillium* strains - including few of the features not allowing to classify them to any of so far known species - were isolated both from the scolytids and from accompanying them mites, but these materials have now been successively elaborated. From the commonly occurring in these materials acaropathogenic species *Hirsutella* cf. *brownorum*, *H. minnesotensis*, *H. nodulosa* and *H. rostrata*, the two latter infected also adult bark beetles, whereas from the larvae and pupae some supposed nematophagous anamorphs were isolated, among them *Harposporium janus* and *Haptocillium* sp.

Key words: arthropod-pathogenic fungi, Entomophthorales (Zygomycota), Hypocreales, (Ascomycota) anamorphs

INTRODUCTION

Entomopathogenic fungi, mostly these infecting numerous species of noxious insects and mites (the latter named recently “acaropathogenic”), have been long considered as having a great potential in biological control of pest arthropod and many of these species were included in different ways into the contemporary Integrated Pest Management (IPM) programs (Pell et al. 2001; Déguine, Ferron 2004; Karg, Bałazy 2009). Despite almost two centuries of rather intensive studies within the taxonomical groups containing fungi pathogenic to arthropods, our knowledge of fungal diversity, ecology and epizootiology is often incomplete and needs particular investigations on their taxonomy, host range and dispersal within ecosystems. Moreover, new species have been described in every year, whereas greatly changing in last decades systematic position of many both higher and lower rank taxonomic sub-units does not facilitate to correctly classify newly discovered forms (Gams 1971; Gams, Zare 2001; Hodge 2003; Hodge et al. 2005; Stensrud et al. 2005; Hibbett et al. 2007; Sung et al. 2007; Rehner et al. 2011).

On the basis of bibliographical data for the European subcontinent, from Austria till the last years of XIX-th century only scanty information on museum collections and occurrence of particular entomopathogenic species – concerning mostly hyphomycetes – were published (Frauenfeld 1849; Bail 1869; Keissler 1924). During last two decades extended studies on hyphomycetes in the soils under various utilization and management systems (Wegensteiner et al. 1998; Hozzank et al. 2003), as well on the Entomophthorales taxa – mainly on aphids (Barta et al. 2003, 2005) – were undertaken and stimulated by the engagement of participants in the activity of COST-Action 842 and IOBC programs.

Studies of these fungi have been relatively well developed in Poland beginning from the second half of XIX-th century and incessantly continued up to the present in several universities and institutes. They have given quite good recognition of the species composition, significance, habitat preferences and natural resources of both main groups of fungal pathogens of arthropods in the Polish countryside (Bałazy, Miętkiewski 2008 and sources cited therein). At the experimental scale several dozen of cordycipitaceous anamorphs of the genera *Beauveria* Vuill., *Isaria* Pers., *Lecanicillium* W. Gams et Zare and *Metarhizium* Sorokin were tested towards their value for application against important pest insects.

The aim of these investigations, carried out within the framework of bi-lateral cooperation project (Scientific and Technological Cooperation: Austria-Poland 12/2006 and 09/2009) in the years 2006-2007 and 2009-2010, was to improve our knowledge and to compare the sets of the fungal pathogens of insects and mites in various ecosystems of Austria and Poland, with particular consideration to forests or semi-natural elements of countryside and suburban areas.

STUDY AREAS

The studies were carried out in Austria and Poland in the years 2006-2007 and 2009-2010 and were conducted as a 2-3 field expeditions during the vegetative season per year (mainly from May to August) in each country. The majority of materials from Austria were collected in the rural and afforested areas of Lower Austria, Upper Austria, Styria and Burgenland. In Poland the greatest part of materials was originated from Podlasie regions, including the Białowieża Primeaval Forests and north-eastern outskirts of the country near Augustów. Relatively rich materials have also been collected in forests, wetlands, orchards and arable fields around westerly situated city of Siedlce in Mazowsze region. In the western part of the country the majority of investigations has been done around the city of Poznań, including the Wielkopolski National Park and in mid-field afforestations of D. Chłapowski's Agro-ecological Landscape Park, situated in the central part of Wielkopolska Region.

MATERIAL AND METHODS

In every area prospected special attention was paid to dead arthropods bearing symptoms of a fungal infection *in situ*. In all cases these cadavers were stored in Petri dishes, and then brought to the laboratory, where macroscopic and microscopic observations were done and attempts to isolate the etiological agent were carried out. For the isolation and identification of the pathogenic fungi representing the anamorphs of Hypocreales, dead arthropods, either mummified or covered by emerging hyphae, were transferred to moist chambers to encourage sporulation. When the mycelium became well developed, a sample or a portion was transferred onto Sabouraud dextrose agar (SDA). In case of entomophthoralean fungi, fungus-killed insects were allowed to discharge primary and secondary spores on glass slides during a stay in a humid chamber. Isolation of entomophthoralean fungi was carried out by means of the "descending conidia" showering method into SDA medium enriched with egg yolk (SDEYA). For microscopic measurements of fungal structures, material was prepared in lactophenol (LP) with aniline blue or/and in aceto-orcein, according to Keller (1987).

Identification of collected fungal species was done by using standard methods described elsewhere (e.g., Keller 1987; Bałazy 1993; Goettel, Inglis, 1997) and in some cases was verified by molecular analyses (PCR). Fungal nomenclature follows Index Fungorum and in case of Entomophthorales also Keller (2007) while the names of the hosts are given after Schaefer (2010) and Szafer et al. (1988), for animals and plants, respectively.

RESULTS AND DISCUSSION

Results from Austria. In total, 47 species of entomopathogenic fungi were found as pathogens of different arthropods in the country. Twenty six entomophthoralean species from different insects and one species from mites were identified and 16 of them are recorded as new to Austria. From among 21 species of anamorphic Hypocreales (Ascomycota) affecting arthropods, 13 species so far have not been known from this country; among them 9 have been recognised as pathogens of mites (Tabs 1 and 2). The species diversity and composition varied in different habitat types as characterized below.

The most frequent species of the entomopathogenic fungi both in agricultural and afforested areas in Austria were the common and usually worldwide distributed cordycipitaceous anamorphs *Beauveria bassiana*, *Isaria fumosorosea* and in areas of this study less frequently *I. farinosa*. The absolute dominant – *B. bassiana* – is particularly frequent in brushwoods and uncultivated orchards at often abandoned possessions in countryside, where next by relatively numerous small beetles of the families *Chrysomelidae*, *Curculionidae* or *Staphylinidae*, this fungus frequently affected also different plant-hopper individuals. Apart from aforementioned hyphomycetous species single adult beetles and larvae or caterpillars covered by the sporulating mycelium of *Metarhizium anisopliae* were also found on other uncultivated fields or on strongly insolated, stenothermal patches of vegetation, usually adjacent to afforested areas. In clearings and thinned out forests its singly specimens occurred also in forest litter, but they have not been met in subcortical feeding sites of cambioxylophagous insects, despite relatively careful searches. Entomophthoralean species have been found in such conditions only sporadically, by one to few individuals in aphid colonies or on single adults of small plant-hoppers and dipterans – most frequently of the family *Sciaridae* or related nematocerous forms.

In the annual cultures on arable fields only single mycosed insects or their residues with the sporulating mycelium of *B. bassiana* were found almost exclusively on grassy roadsides at the maize or cereal crops. In one of numerous maize fields several living beetles of *Diabrotica virgifera* Le Conte were observed close to a trap for these insects and one dead, covered with the sporulating mycelium of *B. bassiana*, was collected next by, on the roadside. Neither aphids nor other pest insects were observed on plant stems or in ears in the quantities that could suggest serious damages leading to significant losses of yields.

On one ripening sunflower field surrounded by cereal cultures with scanty linear belts of trees and few groups of bushes several dozen of adult cecidomyid cadavers infected by *Entomophthora israelensis* were collected. They occurred by 1-3 on the underside of some sunflower leaves. Apart from small brown spots around the points of the midges' insertion to the leaf no other damages were visible on plants and their fructifications. Living individuals of the midges were lacking.

In a perennial culture of alfalfa on the relatively big field (over 10 ha) high mortality indices of the weevils *Sitona* Germar species were well recognized on several 2 x 2 m randomly scattered research plots. The numbers of mycosed beetles and observed living ones were similar, which allow to estimate their restriction as approximately 50 %. This case could be treated as pre-epizootic one, if the mortality

level increased in next months, which could not be checked because of too short period of investigations. However, such a course of epizootics was previously stated in many regions of Poland (Bałazy 2004). On the basis of visible impairment of plants this culture could be estimated as being in very good state, because of low infestation level both by *Sitona* spp. and by other noxious insects. High mortality of weevils in alfalfa caused by *B. bassiana* in the Austrian case is comparable with the results noted in Polish, French and Romanian areas of these plants in late summer and autumn. More exact considerations about the population dynamics and diseases of alfalfa and clover pest insects require a complex treatment, because of particular interference of natural enemies and chemical control measures on injurious effects during the vegetation season.

As the real epizootics can be treated colonies of aphids and coccoid species on leaves of deciduous trees and shrubs, almost entirely overgrown by several *Lecanicillium* and related hyphomycetous species during and after the inundation in Danube River valley near Vienna. The authors had an opportunity to observe these phenomena during the stay in the first decade of August 2006. Among the isolated strains *Acremonium* cf. *potronii*, *L. lecanii*, *L. longisporum*, *L. muscarium*, *Simplicillium lanozoniveum* were identified on scale insects and aphids on accessible leaves of trees and shrubs. In the forest litter and herbaceous layer of vegetation under canopy (apart from relatively common *B. bassiana* and *I. farinosa*) on one research plot of the area of 4 m², two spiders infected by *Gibellula leiopus* and 6 by *G. pulchra* were found as first for Austria belonging to this genus. On unflooded, open patches of overground vegetation *Hirsutella kirchneri*, *H. thompsonii*, *Ramularia ludoviciana* occurred commonly on eriophyid mites on grasses and *H. nodulosa* on *Tarsonemus potentillae* on *Potentilla anserina*. The real diversity of the fungi under discussion during and after such periodical floods seems to be much greater but a limited duration of the stay did not allow for its better recognition. The diversity and richness of the discussed fungi have been raised by spontaneously forming groups or strips of wild vegetation, including roadsides, balks, ditches, ruderal communities, garden-plots and parks, where the mycopathogens of small arthropods – mainly mites, thripids (thysanopterans) and apterygotes – have so far been very superficially recognized, though several dozen of entomophthoraleans and hyphomycetous anamorphs were recorded as affecting insects and mites or other components of mesofauna (Miętkiewski et al. 2000; Hodge 2003; Bałazy et al. 2008b). In almost all samples of grasses, weeds, forest floor plants and garden flowers inspected within this and a number of other studies, individuals infected by fungous pathogens of the genera *Hirsutella* Pat., *Lecanicillium*, *Neozygites* Wiltaczil, *Simplicillium* W. Gams et Zare – often weakly known or even new for science (Miętkiewski, Bałazy 2003; Bałazy et al. 2008a) – were recorded (Tabs 1 and 2) or are awaiting the formal description. A number of quantitative considerations on infection intensity of some more important species of mites indicate often significant differences between particular habitats or even countries. For instance the degree of infection of eriophyid species of the genera *Abacarus* (Nal.) and *Aculodes* Keifer by the representatives of *Hirsutella* species in France were in general considerably lower than in similar sites in Poland (Tkaczuk et al. 2004), whereas first data from similar comparisons from Austria show the values generally lower than in France. On the other hand, the numbers of the mite *Tarsonemus potentillae* infected by *H. nodulosa* on the leaves of *Potentilla anserina* was in Austrian samples two-fold

higher than in Poland. Of course, the changes between particular sites within every country fluctuate also very strongly.

Significant role of natural and semi-natural patches or strips of vegetation has been stressed in many contemporary approaches to the Integrated Pest Management (IPM), in which the landscape diversity – enhancing occurrence and richness of entomophagous components and entomopathogens, among them mostly fungi – gains more and more in value (Chandler et al. 1997; Hozzank et al. 2003; Meyling, Eilenberg 2006; Tkaczuk 2008).

The majority of arthropod mycoses caused by entomophthoralean fungi in Austria was recorded in humid deciduous or mixed forest habitats. Especially interesting is the first record of *Batkoa major* and *Entomophaga tipulae* on *Tipula* species, *Zoophthora crassitunicata* on cantharid beetles, *Z. geometricalis* on adult individuals of Lepidoptera and *Z. cf. radicans* on larvae of *Gastrophysa viridula* (Tab. 1). The latter fungus is well known pathogen of larvae and adult beetles of *G. viridula* feeding on leaves of *Rumex* spp., among the meadows near rivers, but so far known only from locations in eastern Poland. The Austrian strain of this fungus was isolated in June 2010 from larvae of *G. viridula* feeding on dock growing in the forest habitat near Pöllau/Rabenwald (Styria).

Conclusions of Austrian investigations. Excluding several unidentified strains isolated from residues of dead insects and of not verified pathogenic abilities in relation to living insects, 47 entomopathogenic species have been recorded from Austria within the discussed projects. Considering relatively short periods of field researches, this result can be estimated as good beginning material for widened

Table 1
Entomopathogenic fungi collected in the years 2006-2007 and 2009-2010 on insects and spiders in Austria (* – species new for Austria)

Fungus species	Host(s); region (locality); habitat; period and intensity of appearance (epizootic, common, rare, exceptional)
Entomophthorales	
<i>Batkoa apiculata</i> (Thaxt.) Humber	Polyphagous; Lower Austria, Styria; forests and rushes; late spring to autumn; common.
* <i>Batkoa major</i> (Thaxt.) Humber	<i>Tipula</i> L. species; Burgenland; deciduous forest; late spring; rare.
* <i>Conidiobolus cf. adieretus</i> Drechsler	Subcortical Collembola; Lower Austria; forest meadow ecotone, in decayed oak wood; exceptional.
* <i>Entomophaga grylli</i> (Fresen.) Batko	Acridiidae; Styria; meadows; summer; common, on small plots often epizootic.
* <i>Entomophaga tipulae</i> (Fresen.) Humber	<i>Tipula</i> spp.; Lower Austria; deciduous forest; common.
* <i>Entomophthora israelensis</i> Ben Ze'ev et Zelig	<i>Cecidomyiidae</i> ; Burgenland; sunflower leaves; summer; common.
<i>Entomophthora muscae</i> (Cohn) Fresen.	Calyptrate flies; Lower Austria, Styria, meadows, orchards; late spring to autumn; common on single, scattered host individuals.
<i>Entomophthora planchoniana</i> Cornu	Aphids; Burgenland, Lower Austria, wild plants; late spring to autumn; common, locally epizootic.
* <i>Entomophthora schizophorae</i> Keller et Wilding	Calyptrate flies; Upper Austria; late summer; rare.
* <i>Erynia ovispora</i> (Nowak.) Remaud. et Hennebert	Brachycerous flies; Lower Austria; afforested ravine of mountain stream; rare.
<i>Furia sciarae</i> (Olive) Balazy	<i>Sciaridae</i> flies; Lower Austria, Styria; forest; late spring to autumn, common, seldom epizootic.
* <i>Furia vomitoriae</i> (Rozsypal) Humber	<i>Calliphoridae</i> flies; Lower Austria; city park, shelterbelt; summer; rare on single host individuals.

Table 1 – cont.

<i>*Neozgytes parvispora</i> (MacLeod et Carl) Remaud. et Keller	Thrips; Lower Austria; meadow; exceptional.
<i>Pandora dipterigena</i> (Thaxt.) Humber	<i>Sciaridae</i> flies; Lower Austria; deciduous forest, city park; late spring to summer; common.
<i>*Pandora echinospora</i> (Thaxt.) Humber	<i>Lauxaniidae</i> flies; Lower Austria, Styria; deciduous forest, city park; summer; rare.
<i>Pandora neoaphidis</i> (Remaud. et Hennebert) Humber	Aphids; Lower Austria, Burgenland; sunflower fields, shelterbelts; late spring to autumn; common on aphids, locally epizootic for <i>Impatiens asiaticum</i> Nevsky on <i>Impatiens parviflora</i> L.
<i>Pandora</i> sp. (I)	<i>Sciaridae</i> flies, Upper Austria; deciduous forest; late summer; rare (possibly more than one species).
<i>Pandora</i> sp. (II)	An adult chrysomelid beetle (Halticinae) – one individual on the common beech leaf; Vienna municipal forest; exceptional.
<i>Tarichium</i> sp.	<i>Phyllobius</i> Germar species, adult individual; Styria; deciduous forest; late summer; exceptional.
<i>*Zoophthora aphrophorae</i> (Rostrup) Balazy	Planthoppers, Styria, Upper Austria; forest floor vegetation of deciduous or mixed stands; late spring and summer; rare.
<i>*Zoophthora crassitunicata</i> Keller	Cantharid beetle; Upper Austria; deciduous forest; late spring; exceptional.
<i>*Zoophthora geometralis</i> (Thaxt.) Batko	Adult individuals of Lepidoptera sp. div.; Styria; deciduous forest; summer; exceptional.
<i>*Zoophthora opomyzae</i> Balazy et Miętk.	<i>Opomyza florum</i> Fabr.; Lower Austria; under a leaf of <i>Arctium lappa</i> L.; late spring and summer; rare.
<i>*Zoophthora petchii</i> BenZe'ev et Kenneth	Planthoppers; Lower Austria, Styria; deciduous forest; late summer; rare.
<i>*Zoophthora</i> cf. <i>radicans</i> (Bref.) Batko	<i>Gastrophysa viridula</i> De Geer feeding on plants of <i>Rumex</i> L.; Styria; mixed forest; early summer; exceptional.
<i>Zoophthora</i> sp.	An adult individual of Neuroptera; Styria, deciduous forest; early summer; exceptional.
Hypocreales (Ascomycota) – only anamorphs	
<i>*Acremonium potronii</i> Vuill.	Scale insects on leaves; Danube River valley near Vienna; rare.
<i>Beauveria bassiana</i> (Bals.-Criv.) Vuill.	Different insects; Lower and Upper Austria, Burgenland, Styria; whole vegetative season; common.
<i>Beauveria brongniartii</i> (Sacc.) Petch	Larvae of <i>Hylobius abietis</i> L.; coniferous forest; spring; exceptional.
<i>*Gibellula leiopus</i> (Vuill. ex Maubl) Mains	Spiders on <i>Rubus</i> L. species; Lower Austria; deciduous forest; mid-summer; rare.
<i>*Gibellula pulchra</i> Cavara	Spiders on <i>Rubus</i> sp.; Lower Austria; deciduous forest; mid-summer; rare.
<i>Isaria farinosa</i> (Holmsk.) Fr.	Individuals of different insects in the forest litter; Lower Austria; mixed forest, late spring and summer; common but scattered on single host individuals.
<i>Isaria fumosorosea</i> Wize	Larvae and pupae of Lepidoptera in the forest litter and xerothermic deforested strips; often by several host individuals on single plot; Lower Austria; deciduous forest; late summer; common.
<i>*Lecanicillium</i> cf. <i>dimorphum</i> (J. D. Chen) Zare et W. Gams	Sciarid larva; reared subcortical detritus in spruce forest; Upper Austria; late spring; exceptional.
<i>Lecanicillium lecanii</i> (Zimm.) Zare et W. Gams	Scale insects on leaves; Danube River valley near Vienna; deciduous afforestations; epizootic.
<i>*Lecanicillium longisporum</i> (Petch) Zare et W. Gams	Aphids and scale insects on leaves; Danube River valley; Sciarid flies and mites; subcortical beetles' feeding sites on spruce; coniferous forest stands; common.
<i>*Lecanicillium muscarium</i> (Petch) Zare et W. Gams	Adult gnats in reared subcortical detritus; Upper Austria; forest; late spring; obtained mostly in laboratory rearings; aphids; Danube River valley; common.
<i>Metarhizium anisopliae</i> (Metschn.) Sorokin	Beetle larva found on a deforested patch, common in soil and forest litter; Burgenland, Lower Austria, Styria; arable fields, forests, orchards; common but occurring singly.
<i>Tolypocladium</i> sp.	Isolated on selective medium from the forest soil; Styria, deciduous forest; summer; rare.

Table 2

Acaropathogenic fungi collected in the years 2006-2007 and 2009-2010 on phytophagous, predacious and saproxylic mites in Austria (* – species new for Austria, ** – new species)

Fungus species	Host(s); region (locality); habitat; period and intensity of appearance (epizootic, common, rare, exclusive)
* <i>Hirsutella</i> cf. <i>brownorum</i> Minter et Brady	Tarsonemids and juvenile instars of other mites in rearing materials of subcortical insects; Upper Austria; spruce forest; summer; common but usually on single, scattered individuals.
** <i>Hirsutella danubiensis</i> Tkaczuk, Bałazy et Wegenst.	<i>Tetranychus urticae</i> Koch feeding on <i>Potentilla anserina</i> L.; Lower Austria; meadow; late spring and summer; exceptional.
* <i>Hirsutella kirchneri</i> (Rostrup) Minter, Brady et Hall	Eriophyid mites <i>Aceria tussilagofoliae</i> Boczek, <i>Abacarus hystrix</i> Pgst.; Lower Austria, Styria; meadows; late spring to autumn; common.
* <i>Hirsutella nodulosa</i> Petch	<i>Tarsonemus potentillae</i> on <i>Potentilla anserina</i> ; Lower Austria, meadows; late spring to autumn; rare.
* <i>Hirsutella thompsonii</i>	On eriophyid mites feeding on grasses and <i>Eriophyes piri</i> Pgst. in leaves of <i>Pyrus communis</i> L.; Lower and Upper Austria; meadows, pear orchards; summer to autumn; common.
* <i>Lecanicillium</i> cf. <i>dimorphum</i> (J.D. Chen) Zare et W. Gams	<i>Dendrolaelaps</i> Halbert sp. div. and other gamasids; Lower and Upper Austria; in galleries of subcortical insects; late spring and summer; rare.
<i>Lecanicillium</i> cf. <i>lecanii</i> (Zimm.) Zare et W. Gams	Predatory mites in rearing materials of subcortical insects; Upper Austria; spruce bark; summer; rare.
* <i>Lecanicillium muscarium</i> (Petch) Zare et W. Gams	Acarids, gamasids, uropodids; Lower and Upper Austria; in subcortical galleries; late spring and summer; rare.
* <i>Lecanicillium psalliotae</i> (Treschew) Zare et W. Gams	<i>Dendrolaelaps</i> spp.; Upper Austria; in subcortical material; late spring and summer; rare.
<i>Neozygites floridana</i> (Weiser et Muma) Remaud. et Keller	<i>Tetranychidae</i> mites; Burgenland, Lower Austria; <i>Potentilla anserina</i> plants on meadows; late spring to autumn; rare.
* <i>Ramularia ludoviciana</i> Minter, Brady et Hall	<i>Abacarus hystrix</i> Nal., <i>Aculodes mckenziei</i> (Keifer), Lower Austria; on grasses; late summer and autumn; common.
<i>Simplicillium lanosoniveum</i> (J.F.H. Beyme) Zare et Gams	On mites in rearing materials of subcortical insects; Upper Austria; spruce bark; summer; rare.

future studies. On the basis of the authors' experience from long-lasting researches in Poland, neighbouring areas of Southern Bavaria in Germany and Parisian Basin in France, a better recognition of the real abundance and diversity of the discussed groups of fungi requires several repeatable visits during the vegetation seasons, at least through three to five years. The numbers of reported taxa from chosen grasslands and neighbouring wetlands in aforementioned countries were respectively 74, 56 and 88. They were found in Bavaria and France after eight years of two yearly stays, 10 -12 days each (Bałazy 1997, 2006, 2007; Bałazy et al. 2008a).

Results from Poland. The Polish studies within the discussed projects were focused on the interactions between different trophic groups of forest insects and their fungal pathogens in various microhabitats, connected mainly with the feeding sites of saproxylic cambio- and xylophagous invertebrates (Bałazy 2011), and on the possible links between the species composition of this group of fungi and the abundance of foliophagous, saprotrophic and entomophagous arthropods in assimilative apparatus of forest floor vegetation, undergrowth and litter.

Especially interesting is the first record of mycoses (up to 60% mortality), caused by species of the genus *Zoophthora* Batko in a local (on about 0.7 ha of the mixed forest) population of adult *Phyllobius* beetles in Białowieża. From the materials collected in the same habitat in 2009 and 2010 several beetles infected by very rare species – *Pandora phyllobii* (Bałazy) S. Keller – were selected. During our joint research in the vicinity of Siedlce, we found for the first time in Poland and Europe juvenile and adult individuals of earwigs infected by the fungus *Furia* cf. *shandongensis* Wang,

Lu et Li. They were attached to the undersides of the leaves of hornbeam and linden seedlings and of some herbaceous plants of the deciduous forest floor, under the dense canopy of the hornbeam.

In the floor vegetation of two other coniferous forests situated near by Siedlce, a *Zoophthora* species was found and isolated causing up to 30% mortality in whiteflies of the genus *Aleyrodes* L. feeding on *Chelidonium majus* L. With respect to morphology it seems to be very close to *Z. radicans*, but in the light of recent recommendations to verify species identity by molecular markers, this resemblance should be confirmed, all the more so because this is the first case to find a species of *Zoophthora* on whiteflies. Two recently described new species of *Hirsutella* – *H. danubiensis* from Austria and *H. vandergeesti* Bałazy, Miętk. et Tkaczuk from France – were recorded as common in several afforested posts in Siedlce district, the first on tetranychid mites mostly from raspberry, and the second almost exclusively on predaceous phytoseiids – seldom on spider mites – from raspberry and bramble leaves. Moreover, an entomophthoralean species of the peculiar *Apterivorax/Neozygites* features appeared in the materials from raspberries and is actually under elaboration.

The heavy autumnal epizootics in a chalcid wasp of the family Ptomalidae caused by *Zoophthora* cf. *lanceolata* Keller had been quantitatively characterized in the part of afforested area of the Warta River valley in the Wielkopolski National Park near Puszczykowo, where the level of the host-insects' mortality was the most highest. The insects withering away were attached to the underside of undergrowth leaves on the above ground layers up to about 4 m over the forest floor. The first dead individuals appeared on the turn of July/August 2009, in the dense forest undergrowth. Their number increased quickly, attaining 30-50% mortality by the end of August, whereas in mid-September it surpassed 80% and beginning from mid-October only single living individuals were sporadically observed. In two following years the epizootics were heavier and the cases of the disease were found in several other forest stands around the city of Poznań. The entomophthoralean pathogens were rather seldom noted on hymenopteran insect parasitoids, usually as incidentally found on single individuals, so this case merits a more exact treatment. Unexpectedly high diversity has been revealed in the entomophthoroses caused by *Batkoa apiculata* and several *Pandora* Humber and *Zoophthora* Batko in populations of small, mostly nematocerous dipterans, in late spring and autumnal seasons, on the leaves of forest floor and undergrowth vegetation.

Thorough searches showed several new fungal pathogens infecting larvae and pupae of bark beetles *Ips typographus* (L.) and *Dryocoetes villosus* (Fabr.) in sectors of larval tunnels or undamaged cambium entirely isolated from surrounding damaged space. Apart from *Hirsutella* and *Lecanicillium* some isolated from them anamorphs resemble these reported as nematode or tardigrade pathogens. Adult individuals of both aforementioned bark beetles appeared susceptible to infection by *Hirsutella entomophila* Pat. and *H. rostrata* Bałazy et Wiśn., whereas the larvae of sciarid midges died from infection by *H. cf. haptospora* Bałazy et Wiśn. Both the latter belong to the pathogens of mites, whereas *H. minnesotensis* Chen, Liu et Chen – known as the nematopathogenic species – was many times observed on (and isolated from) mites, mostly tarsonemid, occurring in subcortical insects' feeding grounds or from wooden waste mixed with soil. Though the species of the genus *Harposporium* Lohde have been treated as generally nematophagous, *H. janus* Shimazu et Glockling develops

frequently in cerambycid larvae and pupae in dead wood, showing all the signs of fungous disease, but attempts to infect the healthy larvae with sporulating mycelium on mycosed individuals gave positive effect only in two cases. This could suggest some form of dependence of fungal infection on invasive action of nematodes. From subcortical detritus were also isolated other fungi known as potential pathogens of lower invertebrates, as *Haptocillium* W.Gams et Zare sp., *Drechmeria* W.Gams et H.-B. Jansson sp., *Simplicillium* sp. and others very weakly so far recognized, which have to be involved into matter transformation in these specific microhabitats.

Conclusions of Polish investigations. From among 51 species of fungi affecting different arthropods in Poland, 28 species of Entomophthorales and 23 anamorphic Hypocreales (Ascomycota) were separated; among them 13 have been recognised as mite pathogens. Numerous cultures of both clavicipitalean anamorphs and entomophthoralean strains were prepared for the DNA molecular analyses, in order to assess their genetic and taxonomic resemblance. All the above materials are actually in course of preparation for publishing.

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Poszerzone badania nad różnorodnością grzybów – patogenów stawonogów w Austrii i w Polsce

Streszczenie

W latach 2006-2007 oraz 2009-2010 w ramach dwustronnej współpracy (Scientific and Technological Cooperation Project: Austria - Poland 12/2006 i 09/2009), w wybranych siedliskach w Austrii i w Polsce prowadzono badania nad grzybowymi patogenami stawonogów, głównie owadów i roztoczy. W sumie w Austrii stwierdzono występowanie czterdziestu siedmiu gatunków grzybów patogenicznych w stosunku do stawonogów. Wśród nich odnotowano 26 gatunków owadomorków (*Entomophthorales*), z czego 16 to gatunki niewykazywane wcześniej z tego kraju oraz 21 gatunków grzybów reprezentowanych przez anamorficzne formy *Hypocreales* (*Ascomycota*), w tym 13 gatunków nowych dla Austrii. Spośród 51 gatunków grzybów patogenicznych dla stawonogów, które rozpoznano w ramach badań prowadzonych w Polsce, 28 należało do owadomorków, a 23 to anamorfy workowców (*Ascomycota*), z których 13 gatunków wykazuje właściwości patogeniczne w stosunku do roztoczy. Do gatunków grzybów najczęściej spotykanych na roztoczach fitofagicznych oraz występujących w zbiorowiskach owadów zasiedlających drewno, należały gatunki z rodzaju *Hirsutella*, z których żaden nie był dotąd notowany w Austrii. Spośród nich tylko *H. kirchnerii* i *H. thompsonii* wydają się w istotny sposób redukować populację szpecieli w obu krajach. Dwa nowe gatunki *Hirsutella danubienensis* i *H. vandergeesti*, opisane w 2008 roku, są efektywnymi patogenami szkodliwych roztoczy z rodziny przędziorkowatych (*Tetranychidae*), chociaż ostatni z wymienionych gatunków pasożytuje również na roztoczach drapieżnych (*Phytoseidae*). Kilka gatunków owadomorków powodowało epizooce, które wpłynęły na znaczne ograniczenie populacji różnych taksonów stawonogów w obu krajach. Szczególnie godny uwagi jest przypadek wysokiej śmiertelności (do 60% osobników w populacji) chrząszczy z rodzaju *Phyllobius* zaobserwowany w Puszczy Białowieskiej, a spowodowany przez nieopisany dotychczas gatunek grzyba z rodzaju *Zoophthora*. Efektem wspólnych badań jest także odnotowanie po raz pierwszy w Polsce i w Europie, występowania *Furia* cf. *shandongensis* na skorkach (*Dermaptera*) oraz *Hirsutella entomophila* na imagines *Ips typographus* w siedliskach leśnych.