КРАТКИЕ СООБЩЕНИЯ

A COMPARATIVE ANALYSIS OF THE CULTURED MICROMYCETES IN OLIGOTROPHIC PEATLANDS OF NATURAL BIOSPHERE RESERVATIONS LOCATED IN THE NORTHERN AND CENTRAL PARTS OF RUSSIA

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The current study addresses the fungal diversity of the peatlands that vary geographically and geologically, in the central and northern parts of Russia. The central (Smolenskaya area, National Park «Smolenskoye Poozerie») and northern (White Sea Biological Station area) peatlands have a different geological history, the former have a glacial origin, while the latter are of marine origin. Our aim was to investigate the fungal biota of these zones full of *Sphagnum* moss, and possibly reveal the species that are pertinent to these particular habitats.
In both investigated areas, we found an overall poor species diversity with a high fraction of sterile mycelia. Many penicillia species were dominating in all peat samples – *P. thomii*, *P. spinulosum*, *P. glabrum*, *P. funiculosum*, *P. aurantiogriseum* (together with *Geotrichum candidum*), many of them are known to degrade *Sphagnum*. We were particularly interested in the dynamics of the species from the genus *Oidiodendron* that have been shown to be involved in primary *Sphagnum* degradation. The northern area contained a larger species diversity of *Oidiodendron*, as compared to the central zone. Interestingly, insect-associated species of the genus *Tolypocladium* and *Beauveria*, along with the psychrotolerant species of *Geomyces* and *Tolypocladium* were recovered only from the northern peatlands. *Aspergillus* spp. were found in peatlands of the central zone only. We link this result to the climatic features of the area, but also different invertebrate contents that may be utilized by fungi. We detected a tendency of an increasing diversity of *Oidiodendron* species, psychrotolerant and entomopathogenic fungal species as well as a proportion of sterile forms in the northern area.

Key words: entomopathogenic fungi, fungi decomposing *Sphagnum,* fungi in peatlands, micromycetes in peatlands, *Oidiodendron* spp., psychrotolerant fungi, *Tolypocladium* spp.

Oligotrophic peatlands cover more than 10% of Russia's territory and are the largest reservoirs of peat among peatland types in Russia (Yurkovskaya, 2004). Although oligotrophic peatlands are prevalent in northern areas such as taiga and tundra zones, they also occur more southerly, to the extent of the steppe zone. The peat accumulation is caused by microorganisms that decompose organic matter at a relatively slow rate. The slow rate is corroborated by extremely unfavourable abiotic conditions: low oxygen, low temperature, low pH, high humidity and presence of inhibitory phenolic compounds from a local vegetation, mostly *Sphagnum* mosses. Specific groups of fungi and bacteria are the main players involved in the process of organic matter decomposition in peat (Kachalkin et al., 2005; Gilbert, Mitchell, 2006; Thormann, Rice, 2007; Andersen et al., 2010; Golovchenko et al., 2013). However, the diversity of fungi inhabiting oligotrophic peatland remains understudied, especially in the northern regions. The comparison of the fungal biota between oligotrophic peatlands of both central and northern areas would allow to evaluate the fungal species that are shared (or different) among habitats, possibly shedding some light on their physiology in peat. Existing studies suggest that the fungal diversity among oligotrophic peatlands may vary significantly (Thormann et al., 2004; Thormann, Rice, 2007). The reason is believed to be the unique associations of fungi with specific plants, which are pertinent to a given area (Nilsson et al., 1992; Thormann et al., 2004; Filippova, 2015).

Material and Methods

We have been monitoring fungi at the White Sea coastal and Smolensk region peatlands for a number of years now. Using various approaches, we sought to recover most of the culturable fungal species from that area (Bilanenko, Grum-Grzhimaylo, 2007; Grum-Grzhimaylo et al., 2010, 2012, 2016). Since the *Sphagnum* mosses are the main ingredient of peat, we collected and analyzed the samples from the vertical columns of peat deposits where dominated by *Sphagnum* spp. Hence, our comparative analysis of the fungal biota in differently located oligotrophic peats was accounting for the climatic factors (mainly temperature) rather than plant-associated differences of fungal communities. The pH values of the samples in both regions were similar and varied from 4.0 to 4.5. Climatic factors globally affect the distribution of biota, including one of the most prominent inhabitants of the oligotrophic peats – an arctic mosquito (*Aedes* spp., *Culex* spp.). During warm season, these species reproduce in bulk, and along with the plant material, can act as a food source for fungi (Bubnova et al., 2014). Our research attempts to compare fungal diversity of the two distinct following types of peatlands – the White Sea coastal peatlands (White Sea Biological Station, WSBS, northern, 66°34′ N, 33°08′ E) and the Smolensk region peatlands (National Park «Smolenskoye Poozerie», SP, central, 55°53′ N, 31°40′ E) (Fig.). The first type appears to have been formed as a result of a part of the sea becoming isolated in the course of a land uplift, followed by both demineralization and bogging (Shaporenko et al., 2005; Pantyulin, Krasnova, 2011), whereas the central type shows boggy depressions of glacial origin (Smolensk LakeLand…, 2016). The comparison of the fungal biota between the two boggy areas may give us a clue about the shared species, but also unique ones that may contribute to local ecology of a given peatland.

The climate of the SP is moderately continental with well-defined seasons. Wet air masses from the Atlantic release frosts and snowfalls in winter and a high temperature and rain in summer. The average yearly temperature here is about 4.3°C (National Park…, 2016).The climate of the WSBS region is transitional from marine polar to moderately continental. There are long winter and spring, short summer and long autumn at the WSBS. The average yearly temperature here is about 0.5°C (Shilovtseva, 2009). In June 2006 29 and 25 peat samples from three SP peatlands were taken and from three WSBS peatlands in June 2007, subsequently. The detailed description of the peatlands and taking, storing and cultivation of the samples has been exposed in our earlier works (Bilanenko, Grum-Grzhimaylo, 2007; Grum-Grzhimaylo et al., 2016). Only the differences in the methods are remarked here. The peat samples from the SP were stored at deep freeze until the cultivation, whereas the samples from the WSBS

Results and Discussion

Our results indicate that the explored peatlands of both regions show low species diversity of the cultured micromycetes. The analysis of the SP

Fig. Map of both research locations.

samples helped to identify 39 species of fungi and to reveal 3 sterile isolates. The samples of the WSBS peatlands contained 31 species and 8 sterile mycelia. Only 8 fungal species were found in both regions (sterile isolates excluded) (Table). It is known that specific conditions of peatlands restrict the diversity of fungi (Grum-Grzhimaylo, Bilanenko, 2010).

The prevailing number of anamorphic species belonged to *Ascomycota*, mostly *Penicillium* spp. These results do not disprove the existing view on the mycobiota of peatlands (Thormann, Rice, 2007). The following species were found in both regions: *P. thomii*, *P. spinulosum*, *P. glabrum*, *P. funiculosum*, *P. aurantiogriseum.* The capacity of *P. spinulosum* and *P. thomii* to decompose *Sphagnum* was mentioned in a number of earlier mycological works (Chastuchin, 1967; Dickinson, Maggs, 1974). *P. spinulosum* is regarded as a typical species for a humus soil horizon and oligotrophic peatlands (Summerbell, 2005). This species is widely spread in the soils of tundra and taiga and in bogs of various types. *P. spinulosum* is capable to grow in a wide range of temperatures (from 5°C to 42°C). The upper layers of peatlands, including the peatlands of temperate zones, are characterized by drastic changes in temperature regime. *P. thomii* is capable to develop in the temperature range of 5–37°C. It is found in all complexes of filamentous fungi isolated from acidic soils, in peatlands and from bog plants (Domsh et al., 2007). *P. glabrum* is a common species in podsolic and other acidic soils, raised bogs and fens. *P. glabrum* is widely spread in northern regions, including the soils of the Arctic tundra (Nilsson et al., 1992; Domsh et al., 2007). In our research this species was found in two WSBS peatlands and in one SP peatland. The cosmopolitan species *P. funiculosum* (*Talaromyces funiculosus)* was among the dominating species in all SP peatlands (frequency of occurrence was up to 20–30% and the colony-forming units (CFU) value up to 10^4 CFU per 1 gram of a dry sample). *P. funiculosum* was found in all WSBS peatlands and dominated in one of them (frequency of occurrence up to 100%, CFU value up to 10^6 CFU per 1 gram of a dry sample). *P. funiculosum* can grow in a broad temperature range (5–42°C). This species survives in extremely cold and acidic conditions, in low oxygen conditions (Domsh et al., 2007; Grum-Grzhimaylo, 2013). It can be found in all types of soils, in oligotrophic peatlands and in taiga marsh soils. *P. funiculosum* is capable to destroy *Sphagnum* mosses (Dickinson, Maggs, 1974; Thormann et al., 2004; Thormann, Rice, 2007).

We found *Geotrichum candidum* (*Diplodascus geotrichum*) in both peatland systems mentioned in this work. This fungus is also known as a destructor of *Sphagnum* mosses (Dickinson, Maggs, 1974; Karunen, Kalviainen, 1985). It can exist in highly moist conditions (Dix, Webster, 1995). In further research we managed to isolate it in all WSBS peatlands samples we explored.

Oidiodendron species are known as primary destructors of *Sphagnum* mosses (Tsuneda et al., 2001). *Oidiodendron* spp. grow at acidic and even extremely acidic conditions (pH-values from 1.5 to 6.0) (Gross, Robbins, 2000; Domsh et al., 2007). In our research *O. griseum* and *O. periconioides* were found in northern peatlands, *O. cereale* was found in peatlands of the central zone. *O. griseum* was one of the dominant species in WSBS peatlands. *O. griseum* is a typical species of the ericales rhizosphere, peat and podsolic soils**.** *O. periconioides* and *O. cereale* are also known as typical species for *Sphagnum* bogs (Thormann, Rice, 2007). In frost peatlands of the Kola Peninsula we discovered even greater *Oidiodendron* species diversity than in WSBS peatlands (our unpublished data).

The psychrotolerant species *Geomyces pannorum* (*Pseudogymnoascus pannorum*) was identified in WSBS samples with high frequency and abundance. However, it was not revealed in SP samples at all. *G. pannorum* is a psychrotolerant species and it can be found predominantly in northern soils of tundra and in conditions of the natural cryopreservation (Tosi et al., 2002; Kochkina et al., 2007). This species is known as polyextremotolerant, capable to develop under the influence of numerous stress factors such as low temperature, low water activity and lack of oxygen. This allowed it to survive in anaerobic conditions, which are common for cryopegs in permafrost (Kochkina et al., 2007; Shcherbakova et al., 2010). *G. pannorum* can grow in a wide range of pH values (3.5–8.0) (van Oorschot, 1980). This species was isolated in large quantities (CFU value up to 10^5 for 1 gram of a dry sample) at the depth of 1 metre from a peat sample of a northern bog, which supports the conditions of low temperatures and low oxygen content. Psychrotolerant type of temperature adaptation is also common for *Tolypocladium* spp. (Bisset, 1982). *Tolypocladium* spp. were found in the WSBS samples, but not in the SP ones.

Tolypocladium spp. and *Beauveria* spp. (*T. inflatum*, *T. geodes*, *B. bassiana*) are known as associated with insects. These species were found in the northern peatlands only. This fact was also

Table. Fungal species isolated from the peatlands (№№ 1, 2, 3). SP – the National Park «Smolenskoye Poozerie». WSBS – the White Sea Biological Station. Common species for SP and WSBS are in boldface

Species	SP			WSBS		
	1	2	3	1	2	3
Acrodontium crateriforme (J.F.H. Beyma) de Hoog	–	–	–	+	–	–
Alternaria alternata (Fr.) Keissl.	–	+	–	+	–	–
Aureobasidium pullulans (de Bary & Löwenthal) G. Arnaud	+	+	+	+	–	–
Aspergillus fischeri Wehmer	–	+	–	–	–	–
Aspergillus fumigatus Fresen.	–	+	–	–	–	–
Aspergillus niger Tiegh.	+	–	–	–	–	–
Beauveria bassiana (Bals.-Criv.) Vuill.	–	–	–	–	+	–
Botrytis cinerea Pers.	–	–	–	+	–	+
Cadophora fastigiata Lagerb. & Melin	+	–	–	–	–	–
Cladosporium cladosporioides (Fresen.) G.A. de Vries	–	–	–	+	–	+
Cladosporium herbarum (Pers.) Link	–	–	–	+	+	+
Clonostachys rosea (Link) Schroers, Samuels, Seifert & W. Gams	+	–	–	–	–	–
Diplodascus geotrichum (E.E. Butler & L.J. Petersen) Arx	+	+	–	+	–	–
Gibellulopsis nigrescens (Pethybr.) Zare, W. Gams & Summerb.	–	–	–	+	–	–
Lecanicillium evansii Zare & W. Gams	+	–	–	–	–	–
Lecanicillium sp.	–	–	–	–	+	–
Mucor hiemalis Wehmer	+	+	+	–	–	–
Mucor plumbeus Bonord.	+	–	–	–	–	–
Oidiodendron cereale (Thüm.) G.L. Barron	+	–	–	–	–	–
Oidiodendron griseum Robak	–	–	–	+	+	–
Oidiodendron periconioides Morrall	–	–	–	–	+	–
Paecilomyces divaricatus (Thom) Samson, Houbraken & Frisvad	–	+	+	–	–	–
Penicillium aurantiogriseum Dierckx	+	+	+	–	+	–
Penicillium brevicompactum Dierckx	–	–	–	+	+	–
Penicillium chermesinum Biourge	+	–	–	–	–	–
Penicillium citreonigrum Dierckx	–	–	–	+	+	–
Penicillium citrinum Thom	–	+	–	–	–	–
Penicillium dierckxii Biourge	+	+	+	–	–	–
Penicillium dipodomyis (Frisvad, Filt. & Wicklow) Banke, Frisvad & S. Rosend.	+	+	–	–	–	–
Penicillium glabrum (Wehmer) Westling	+	–	–	+	+	–
Penicillium implicatum Biourge	–	–	–	–	–	+
Penicillium lividum Westling	–	+	–	–	–	–
Penicillium nalgiovense Laxa	–	–	–	–	+	–
Penicillium oxalicum Currie & Thom	–	+	–	–	–	–
Penicillium purpurascens (Sopp) Biourge	+	–	–	–	–	–
Penicillium restrictum J.C. Gilman & E.V. Abbott	–	–	–	+	–	–
Penicillium rolfsii Thom	–	–	–	–	+	–
Penicillium spinulosum Thom	+	+	+	+	+	+
Penicillium thomii Maire	–	+	–	+	+	+
Penicillium velutinum J.F.H. Beyma	+	–	–	–	–	–
Penicillium vinaceum J.C. Gilman & E.V. Abbott	–	–	–	+	+	+
Penicillium waksmanii K.M. Zaleski	+	+	–	–	–	–
Pseudogymnoascus pannorum (Link) Minnis & D.L. Lindner	–	–	–	+	–	–
Phialophora europaea de Hoog, Mayser & Haase	–	+	–	–	–	–
Rhizopus stolonifer (Ehrenb.) Vuill.	+	–	–	–	–	–
Sclerotinia sp.	–	–	–	+	+	–

The end of a Table

Species	SP			WSBS		
	1	2	3	1	2	3
Talaromyces aculeatus (Raper & Fennell) Samson, N. Yilmaz, Frisvad & Seifert	–	+	–	–	–	–
Talaromyces diversus (Raper & Fennell) Samson, N. Yilmaz & Frisvad	–	–	–	–	+	–
Talaromyces funiculosus (Thom) Samson, N. Yilmaz, Frisvad & Seifert	+	+	+	–	+	+
Talaromyces purpureogenus Samson, Yilmaz, Houbraken, Spierenb., Seifert, Peterson, Varga & Frisvad	+	–	–	–	–	–
Talaromyces rugulosus (Thom) Samson, N. Yilmaz, Frisvad & Seifert	+	–	–	–	–	–
Talaromyces variabilis (Sopp) Samson, N. Yilmaz, Frisvad & Seifert	+	+	–	–	–	–
Talaromyces verruculosus (Peyronel) Samson, N. Yilmaz, Frisvad & Seifert	–	–	–	+	–	–
Thyronectria cucurbitula (Tode) Jaklitsch & Voglmayr	–	–	–	+	–	–
Tolypocladium geodes W. Gams	–	–	–	–	+	–
Tolypocladium inflatum W. Gams	–	–	–	+	–	–
Trichoderma koningii Oudem.	–	–	+	–	–	–
Umbelopsis ramanniana (Möller) W. Gams	+	–	–	–	–	–
Sterile isolates (12 morphotypes)	3	–	–	3	5	1

demonstrated in our later works (Grum-Grzhimaylo et al., 2016). One of the possible reasons for such abundance of *Tolypocladium* spp. and *B. bassiana* in the northern peatlands may be connected with considerable populations of invertebrates, including larvae of sanguivorous mosquitoes, on which these species can grow.

Aspergillus spp. (*A. fischeri*, *A. fumigatus*, *A. niger*) known as typical in the southern regions were found in SP peatlands only.

A considerable share in fungi societies of both regions is represented by sterile mycelia. Their number is higher in northern regions. Using molecular methods we showed later that the majority of sterile forms are basidiomycetes (Grum-Grzhimaylo et al., 2016).

The fact is that the absence of some species in the samples could be explained by the low number of samples or by other methodological errors. For example, the absence of *Trichoderma* spp. in WSBS samples was quite surprising. In the following years we consistently discovered *T. piluliferum*, *T. polysporum, T. harzianum, T. viride* in WSBS peatlands.

We assume that the detected tendency of the increasing diversity and the abundance of *Oidiodendron* spp., psychrotolerant and entomopathogenic fungal species (*Geomyces pannorum*, *Tolypocladium* spp., *Beauveria* spp.) as well as the proportion of sterile forms in the northern area is clear.

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