Microscopic fungal assemblage colonizing deadwood in the Karkonosze and Tatra Mountains

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DOI: 10.24131/3247.170103

Summary:

The aim of the study was to determine the species composition of microscopic fungi colonizing dead wood of birch, beech, sycamore maple and sorb (Karkonosze Mountains), and black alder, mountain pine, willow, Swiss pine, sorb, sycamore maple and birch in the Tatras. The mycological analysis was conducted in 2015 and 2016 in the foothills and in the lower and upper montane zones of the Karkonosze Mounts, Karkonosze National Park, and in 2016 in the Białka Valley and Chochołowska Valley in the Tatras, Tatra National Park. For both, Karkonosze Mounts and Tatras, the dominant microscopic fungi inhabiting dead wood belonged to the genus *Trichoderma*. The Mycobiota of dead wood in the lower classes of decomposition was characterized by a higher species biodiversity but the lower numbers of colonies isolated.

Key words: dead wood, microscopic fungi, decomposition, Karkonosze Mts., Tatra Mts

received: 14.12.2016; accepted: 28.02.2017; published: 18.04.2017

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Introduction

Deadwood – as a complex of dynamically changing microhabitats – is an important element that affects the entire forest ecosystem (Pyle and Brown, 2002; Staniaszek-Kik, 2014). Depending on the tree species and the degree of decay remaining wood necromass modifies the abiotic environment, contributing to the formation of mosaic microrelief. It has a huge impact on the modification of local sunlight, moisture accumulation and buffering temperature and moisture changes. These changes favourably affect the biological activity contributing to its development (Solon, 2003).

Residual necromass is an important reservoir of common, rare and protected species influencing the growth of species diversity (Bartnik, 2007). Groups especially related to the specific microhabitat of deadwood are bryophytes, fungi, lichens, slime moulds and invertebrates (Staniaszek-Kik, 2014). Wood with varying degrees of decay contributes to the development of small vertebrates: mammals and birds. Examples include woodpeckers, which have high demands on the degree of decay and volume (trunk circumference and height) of the accumulated necromass. Based on the inventory of bird species from the group of secondary cavity nesters we can assess the degree of decay of the matter left in

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the forest and the approximate size of dead trees. This is due to differing preferences in the selection of sites for nesting and foraging by different species (Bunnell et al., 2002; Ciach, 2011). There is also stressed the predisposition of the occurrence of selected species of fungi and lichens in forest ecosystems of varying intensity of use. By identifying species mycobiota it is possible to determine the degree of naturalness of the ecosystem and the following changes in biocenoses (Sippola et al., 2000; Czyżewska and Cieśliński, 2003). Fungi, as one of the few organisms decomposing plant cell walls, actively participate in the process of wood decay. They serve an invaluable role in the release of the micro and macro elements concentrated in the wood, needed for the development of biocenoses (Kwaśna et al., 2016).

Dieback of trees is often caused by pathogenic sac fungi. They utilize carbohydrates, proteins and fats. They are isolated from live and dead tissues in tree, as well as insect involved in the spread of fungi. With the decreasing abundance of the environment in easily absorbed compounds, saproxylic and saprophytic fungi occur in the presence of wood. Many of them decompose structural components of wood (cellulose, hemicellulose, lignin) unavailable to other microorganisms. This specific form of decay is led by *Basidiomycota* division. They are accompanied by bacteria and *Ascomycota* form of fungi, which: (i) have the ability to degrade wood components (among the others, representatives of *Xylariales* family with *Daldinia*, *Hypoxylon* and *Xylaria* genera) (ii) are not capable of wood decay and draw nutrients from the products of wood half-decay and metabolites of associated fungi. They all serve as decomposers: decompose and as a result disintegrate organic matter and increase the amount of inorganic matter in the environment. Also mycorrhizal fungi find their niche in the final stages of wood decay (Stokland et al., 2012; Sour et al., 2016; Sour et al.,2016b). 20% of fungi in dead tree trunks are mycorrhizal fungi (Franklin et al., 1981). Improving the conditions for tree growth (especially conifers), they contribute to the success of natural regeneration of forests and vegetation development on new positions.

The role of saprophytes in forest ecosystems, including *Ascomycota* phylum, is often underestimated. While decomposing wood, they participate in the circulation of matter in the nature, and contribute among the others to the creation stable humus, stabilization of pH, increase of water capacity and improvement of the lumpy structure of soil. By shaping habitat conditions preferred by different types of plants and animals, they increase immunity and durability of forests and prevent fires. They also contribute to improving forest health through the production of valuable secondary metabolites (antibiotics, enzymes and vitamins) and encouraging the development of predatory insects and spiders, which are natural enemies of harmful insects (Bartnik, 2007).

The topic of wood decay caused by fungi has been raised so far by many authors. Particularly noteworthy are the studies of Savory (1954), Seifert (1983), Crawford et al. (1990), Worrall, et al. (1997), Harju et al. (2001), Venäläinen et al. (2003) and Fukasawa et al. (2009, 2011). In recent years, the European Parliament has stressed the importance of necromass in shaping forest ecosystems. It recommended member countries of the European Union to take appropriate measures to maintain optimal levels of deadwood in forests (Anon 2011).

The aim of the conducted studies was to determine the species composition of microscopic fungi colonizing birch (*Betula* L.), beech (*Fagus* L.), sycamore (*Acer pseudoplatanus* L.) and rowan (*Sorbus* L.) deadwood in the Karkonosze Mountains and the black alder (*Alnus glutinosa Gaertn*.), mountain pine (*Pinus mugo Turra.),* willow (*Salix* L.), Arolla pine (*Pinus cembra* L.), rowan (*Sorbus* L.), sycamore (*Acer pseudoplatanus* L.) and *birch* (*Betula* L.) deadwood in the Tatra Mountains.

Methodology of the researches

Mycological analysis included birch (*Betula* L.), beech (*Fagus* L.), sycamore (*Acer pseudoplatanus* L.) and rowan (*Sorbus* L.) deadwood in the Karkonosze Mountains. In contrast the material for the research in the Tatra Mountains was black alder (*Alnus glutinosa Gaertn*.), Mountain pine (*Pinus mugo Turra*.), Willow (*Salix* L.), Arolla pine (*Pinus cembra* L.), rowan (*Sorbus* L.), sycamore (*Acer pseudoplatanus* L.) and birch (*Betula* L.) deadwood.

Samples were taken three times during the growing season, i.e. in the spring (April/May), summer (June/July) and fall (October/November) of 2015-2016 in the Karkonosze Mountains and of 2016 in the Tatra Mountains. Research posts in the Karkonosze Mountains in 2015 included three areas of the lower and upper montane of the western districts of the Karkonosze Mountains National Park: Chojnik, Szrenica and Kocioł Szrenicki. In 2016 the posts were located in the foothill areas (Góra Żar and Szerzawa), the lower and upper montane of the Karkonosze Mountains National Park (Petrovka, Wilcza Poreba, Kocioł Łomniczki, Łomniczka Valley) (Table 1, Figure 2). No mycological research of sycamore deadwood was carried out dur-

Fig. 1. Beech deadwood in the Karkonosze Mountains

Source: own elaboration

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ing the second year of the research. Research posts in the Tatra Mountains were located in the Białka and Chochołowska Valley (Table 1). In addition, one post was located in the vicinity of the TPN Nature Education Centre in Zakopane (Figure 3).

Field researches were determining the degree of wood decay by Pyla and Brown scale (2002); and then 10 ca. 5-cm-long pieces of wood were collected with a sterile tool from a depth of about 1-2 cm of one of the selected objects (coarse woody debris or standing deadwood) on 1 monitoring post to determine their colonization by microscopic fungi. The surface of the wood pieces was then flushed with a 1% sodium hypochlorite solution for 5 seconds. The next step was laying six pieces of ca. 0.5 cm-long wood fragments onto each Petri dish with solidified and acidified PDA (potato glucose agar) medium. Fungi growing out of the fragments of wood were cleaved onto PDA slants and identified to the species based on morphological characteristics (construction of the mycelium, the shape and dimensions of the spores, the shape and dimensions of conidiophores, sporulation forms and others) using available keys (Pitt and Hocking, 2009; Watanabe, 2011).

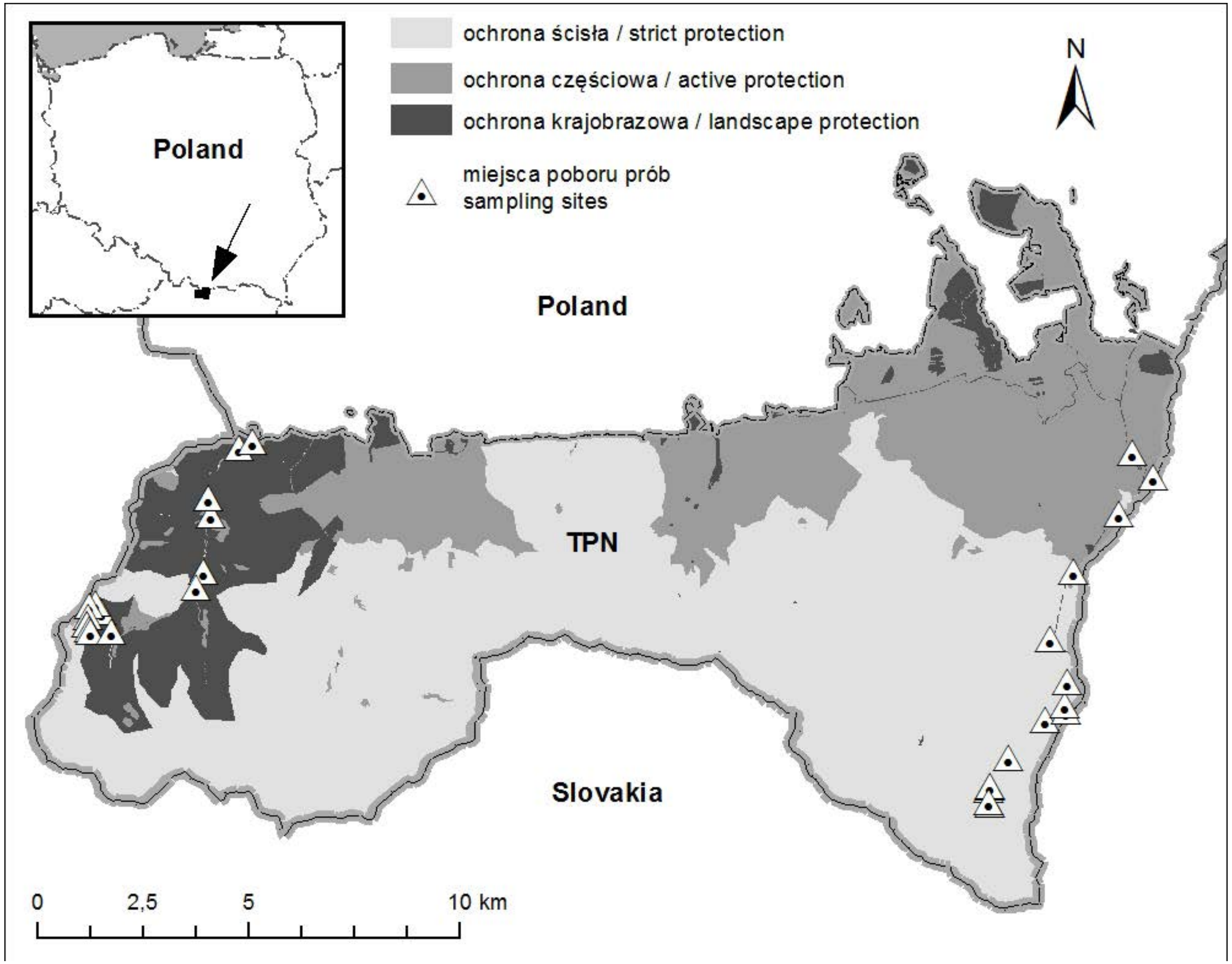

Fig. 2. Distribution of research posts in the Karkonosze Mountains National Park

Source: elaboration of Marcin Wierzbicki, the Karkonosze Mountains National Park

Findings

In 2015 a total of 11 species of fungi were isolated in the Karkonosze Mountains, whereby *Trichoderma harzianum* from the wood of all tree species researched (mountain ash, maple, beech and birch) on all surveyed positions (the Mumlawski Peak, Szrenica, Kocioł Szrenicki and Chojnik). In turn, the second dominant species in mycocenosis of deadwood, *T. polysporum*, was not isolated only from maple deadwood. More species of fungi were identified in the lower grades of wood decay, and some of them, e.g. *Botrytis cinerea* and *Fusarium culmorum*, did not inhabit more decayed wood. 7 species of fungi in a total of 39 isolates were detected on birch deadwood of 2nd degree decay; while in turn: in maple deadwood of 5th degree only one species of fungus (80 isolates); in rowan deadwood of 4 degree decay – 4 species of fungi (67 colonies); in birch deadwood – 4 species (68 isolates) with clear predominance of *T. harzianum* (Table 2).

A total of 19 species of fungi were isolated from deadwood in the second year of studies conducted in

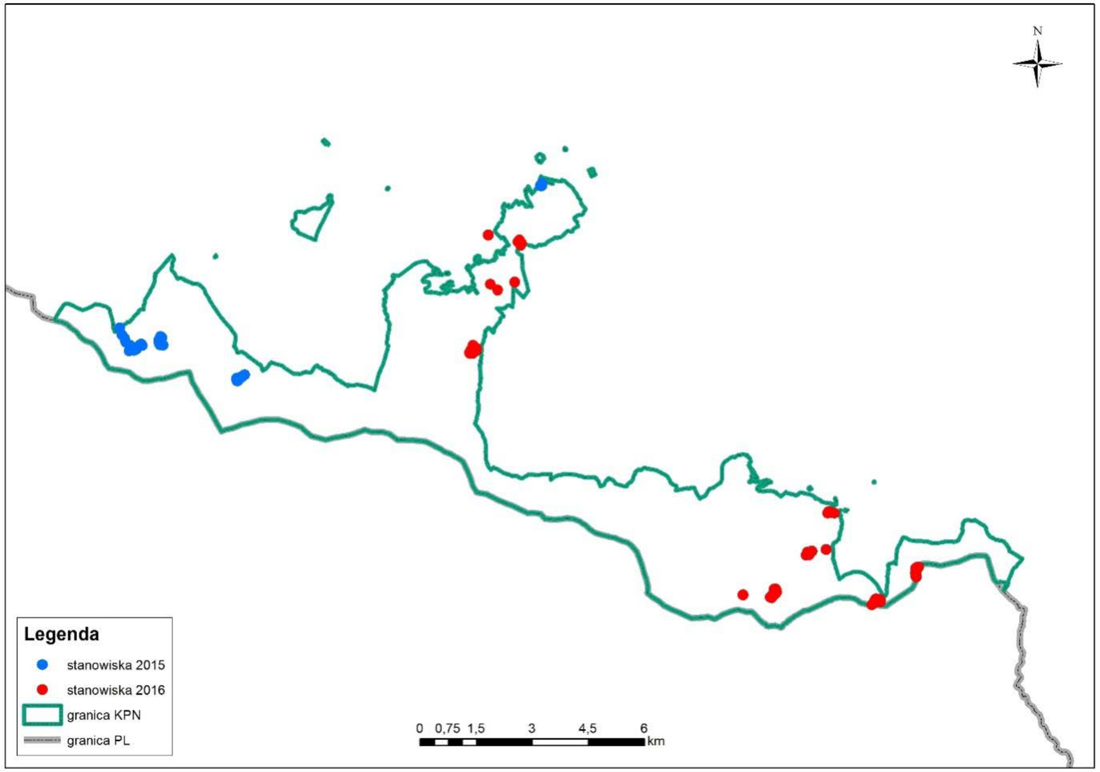

Fig. 3. Distribution of research posts in the Tatra National Park

Source: own elaboration

Fungal species	Deadwood / decay class				
	beech 3	birch 2	mountain ash 4	sycamore 5	birch 4
Alternaria alternata		3			1
Alternaria botrytis			1		
Aspergillus niger	3				
Botrytis cinerea	20				
Fusarium culmorum	1	2			
Fusarium oxysporum		1			1
Mucor hiemalis		1			
Penicillium notatum	15		12		
Trichoderma harzianum	30	12	37	80	63
Trichoderma polysporum	6	10	17		3
Trichoderma hamatum	3	10			
Total	78	39	67	80	68

Tab. 2. Species composition of microscopic fungi isolated from deadwood in the Karkonosze Mountains in 2015 (average number of colonies)

Source: own elaboration

the Karkonosze Mountains. It has been found that contrary to birch wood and rowan most fungi of the *Trichoderma* genus were isolated from beech deadwood. The biggest share in mycocenosis of beech deadwood had species of *T. harzianum* and *T. polysporum*. Beech wood can be characterized by greater biodiversity of mycocenosis than birch and rowan deadwood. 15 species of fungi were isolated from beech wood in spring, whereas this value for birch species totalled 5 and for rowan – 10 (Table 3).

Upon analysing species composition of fungi present in different species of deadwood in the Tatra Mountains, it can be said that most species and colonies were found in rowan deadwood; 5 species and 37 colonies, respectively. Five species of fungi were isolated also from willow wood with respectively fewer colonies. Fungi of the *Trichoderma* genus were not only isolated from Swiss pine deadwood, but unlike the Karkonosze Mountains, *Trichoderma spp.* were not dominants (Table 4).

Discussion

Based on the achieved results it was concluded that the fungi of the *Trichoderma* genus – *T. harzianum* and *T. polysporum* – have the highest share. Fungi of the *Trichoderma* genus are commonly present in the environment and are capable of very rapid colonization of deadwood (Cardoza et al., 2006). They can be found in soil and are responsible for the decomposition of organic tissue (Grondona et al., 1997). Deadwood is thus a kind of reservoir of fungal isolates, which can then be used in various industries (Bartnik, 2007). Fungi of the *Trichoderma* genus may be used in a biological protection of plants against diseases caused inter alia by fungi of the *Fusarium* genus (Grondona et al., 1997, Błaszczyk et al., 2014). Fungi were also observed at the posts in the Karkonosze Mountains and the Tatra Mountains. They could be found on wood by accident or also take part in the so-called mildew wood decay (Sour et al., 2016b). They can be the source of many diseases of plants, both cultivated and wild, but in this case they behaved like typical saprotrophs, decomposing dead organic tissue (Wagah and Muthomi, 2007).

It was found that in the lower classes of decay, wood is populated by a larger number of species, often other than those inhabiting deadwood in the higher classes of decay. This found confirmation in the studies of other researchers (Oszako, 2004, Czekaj and Smith, 2010). The same authors also emphasize the fact that the role of microscopic fungi in the decay of deadwood it is not known to the very end. They probably „open" the door for other groups of fungi and other organisms responsible for the process of decay of deadwood (Sour et al., 2016).

Particularly noteworthy is the fact that it a large number of both species and colonies were found on beech deadwood in the Karkonosze Mountains. Increasing the share of beech population increases the biological activity of the soil, stopping unfavourable process of podsolization (Kabbalah et al., 2013). Thus finding so extensive colonization of the beech deadwood by fungi can prove their positive role in shaping the future of natural regeneration of this species in the Karkonosze Mountains.

Conclusion

We found in this study that the dominant microscopic fungi inhabiting deadwood in selected areas of the Karkonosze and Tatra Mountains are fungi of the *Trichoderma* genus, mostly *T. harzianum* and *T. polysporum*. Lower degree of wood decomposition indicates a much larger variety of species of microscopic fungi than in the higher classes of decay. Some diversity of the mycocenosis of deadwood was also found, depending on the type of wood and its degree of decomposition.

Acknowledgments

The research was funded from the resources of the Forest Fund of the „State Forests" National Forest Holding transferred to the Karkonosze National Park in the years 2015-2016 and the Tatra National Park in 2016.

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