Abstract

Fungi are an important and exceptionally diverse component of microbial communities, and are widely recognized as a fundamental component of ecosystems biodiversity. Up to now, the majority of global fungal diversity remains still unrecognized and the structure of fungal communities in different forest ecosystems is unevenly studied. Unlike the numerous works concerning the effect of management on the forest mycobiome, only a few studies address how fungi from different trophic groups recover from natural and anthropogenic disturbances and develop structural features typical of unmanaged old-growth forests. To recognize how the communities of fungi from different trophic groups are differentiated between forest reserves and managed forests, fungal diversity has been evaluated in three pairs of formerly managed forest reserves established 40–60 years ago ("Bażantarnia", "Olbina", and "Czaplowizna") and forests under standard forest management (Forest Districts Przytok, Kalisz, Łochów) located in continental mixed coniferous forests, recognized as one of the most widespread forest associations in Poland. Most forest tree species in temperate forests form obligatory mutualistic symbioses with ectomycorrhizal (ECM) fungi. Therefore ECM assemblages were analyzed by integration of sporocarp and molecular analyses. Additionally, to evaluate whole soil fungal assemblages from different trophic groups, a new generation metagenomic approach was applied.

The aims of my study were: (i) to describe the soil fungal assemblages represented by different trophic groups in forest reserves and managed stands by using high-throughput Illumina MiSeq sequencing of fungal ITS1 amplicons; (ii) to determine the diversity of ECM fungi based on two different methodological approaches (sporocarp surveys and molecular identification of ectomycorrhizas); (iii) to check whether the studied sites constitute potential habitats for the occurrence of endangered, rare, red-listed, and protected fungi.

In all sites, three pairs of forest reserves and managed forests were established. In each pair of stands, four plots were selected (400 m^2 , $8 \times 50 \text{ m}$). In total, 24 plots were examined: 12 plots located in managed forests and 12 in forest reserves. To evaluate the belowground ectomycorrhizal communities, soil cores were collected five times (in May and October 2015 and 2016, as well as in October 2017). The sporocarp surveying was conducted in 2015, 2016, and 2017 within each sampling plot between May and November. Each plot was visited five times per year. To perform the metagenomic analyses soil samples were collected in September 2018.

The combination of three research methods allowed the identification of 674 fungal taxa from different trophic groups. Among them, 80% fungal taxa were shared between both management types. Formerly managed forest reserves and forests under standard forest management appeared to be similar in terms of total fungal species richness (603 and 607 taxa, respectively). There was no significant

difference between the mean number of taxa per plot and diversity (based on Shannon index) between the management type. A slightly higher number of exclusive fungal taxa was found in managed forests than in forest reserves (71 and 67 taxa, respectively). Among them predominated rarely recorded taxa, usually found in single plots. Two- way analysis of similarities revealed that both site and management strategy significantly influenced the fungal species composition, with the site being a primary effect. High-throughput sequencing, sporocarp surveys, and molecular identification of ectomycorrhizas allowed to identify in total of 171 ECM fungal taxa, with 139 in forest reserves and 145 in managed forests. Apart from the shared taxa, each of the identification methods brought a number of taxa characteristic only for particular methodological approach. Therefore, to obtain a comprehensive resolution of the ECM fungal diversity, varied methods should be applied.

Soil fungal communities (altogether from different trophic groups and separately for ECM fungi and saprotrophic fungi) were influenced by dissimilar sets of environmental factors. The volume of coarse and very fine woody debris and soil pH significantly influenced the ECM fungal community, whereas saprotrophic fungi were influenced primarily by the volume of coarse woody debris and soil nitrate concentration. Both, above and below ground structure of communities ECM fungi were shaped by slightly different environmental factors, but generally, soil nitrate concentration and the number of trees were found to be the main drivers shaping the ECM fungal communities.

The three research methodological approaches revealed 22 fungal species of conservation concern (9 ECM fungi and 13 saprotrophic fungi) and three ECM fungal species new to Poland (*Cortinarius holoxanthus, C. lignicola* and *Entoloma boreale*). Additionally, two wood-inhabiting fungi (*Hymenochaete cruenta* and *Hericium flagellum*) strongly connected with silver fir wood were investigated in terms of their distribution in Poland. For *H. flagellum* the habitat preferences and models of the potential range of the occurrence of this fungus and its plant partner, i.e. silver fir were investigated, taking into account climatic niches suitable for the occurrence of these species.

Our results suggest that the transformation of fungal diversity at studied sites after cessation of forest management is rather slow, and both forest reserves and managed forests help uphold fungal diversity. It seems necessary that the research should be repeated at the same sites (especially in the studied reserves) for the next several dozen years. This would allow capturing further changes in the structure of fungal communities associated with the cessation of forest management activities, because, as shown by other studies, it takes an average of 90 years to restore the original state of ECM fungal diversity disturbed by forest management.