Phyton (Horn, Austria) Special issue: "Bioindication ..."	Vol. 36	Fasc. 3	(115)-(120)	15.09.96

Types of Ectomycorrhizae and Mycobioindication of Forest Site Pollution

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Key words: Mycobioindication, types of ectomycorrhizae, forest site pollution.

Summary

KRAIGHER H., BATIČ F. & AGERER R. 1996. Types of ectomycorrhizae and mycobioindication of forest site pollution. - Phyton (Horn, Austria) 36 (3): (115) - (120).

Types of ectomycorrhizae on Norway spruce were determined in soil cores from two differently polluted forest research plots from the emmission zone of the Šoštanj Thermal Power Plant (TPP). All fine roots and ectomycorrhizal root tips were counted and the percentages of different types were calculated. Additionaly, soil and mycorrhizae from underneath fungal fruitbodies were collected in order to characterize new types of ectomycorrhizae by classical anatomical and by molecular methods. 17 types of ectomycorrhizae were determined on 28443 root tips in soil cores from the polluted plot (Zavodnje). On the less polluted plot (Mislinja) 24 different types were determined on 38502 root tips in equal volume and number of soil cores. In the discussiion of mycobioindication of forest site pollution we employed a supposedly pollution sensitive (*Hydnum rufescens*), and supposedly unsensitive (*Paxillus involutus*) fungal species in ectomycorrhizae. However, further screening of comparable forest sites differently influenced by pollution is needed to confirm the choice of species.

Introduction

Mycorrhizae represent the main spatial and temporal linkage between different constituents in a forest ecosystem (DIGHTON & BODDY 1991). Their functional compatibility, physiology and tolerance to different stress factors is species specific (HARLEY & SMITH 1983, GIANINAZZI-PEARSON 1984). Therefore knowledge on the ectomycorrhizal types - species composition and their abundance

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is necessary in order to understand the functioning of a forest ecosystem. An increasing number of reports have been concerned with a decrease in species diversity and the abundance of sporocarps of macromycetes in Europe (ARNOLDS 1991, JAENIKE 1991 etc.). FELLNER 1989 founded a myco-bioindication method of forest site pollution distinguishing three phases of the impoverishment of mycobiocenosis. ARNOLDS 1991 has tested FELLNER's phases and suggested an adaptation of his qualification of impoverishment in respect to different sensitivities of different ectomycorrhizal species.

The long-lasting tolerant species may vary in different forest communities and regions. FELLNER 1989, from observations in Krkonoše, suggested that the fungus *Russula mustelina* might be used as a bioindicative mycorrhizal species for the estimation of forest soil pollution by acid depositions. However, the occurrence of fruitbodies depends on a range of climatic factors in different years, while ectomycorrhizal types are supposed to be present in the soils at any time of the year. Furthermore, a range of ectomycorrhizal fungi, such as Fungi Imperfecti, do not produce any sporocarps. As a result, more detailed studies of ectomycorrhizal potential of forest sites by ectomycorrhizal types determination were proposed, with a view to applying the results to bioindication.

Our study was concentrated on determining types of ectomycorrhizae on two differently polluted forest research plots and in comparing of their occurrence in soil cores throughout the vegetation season.

Materials and Methods

Types of ectomycorrhizae on Norway spruce were determined in soil cores from two differently polluted forest research plots in the emmission zone of the Šoštanj Thermal Power Plant (TPP). The two plots (850 m/asl, distric cambisol, *Luzulo - Fagetum*, predominant tree species *Picea abies*, ca 80 - 100 years old) were as similar regarding site characteristics as it was possible to select, but differently polluted. The plot in Mislinja is relatively unpolluted and the plot in Zavodnje is the polluted one (BATIČ & KRALJ 1990). Soil analysis data are shown in Table 1.

Tab. 1. Sieved soil analysis data for two forest research plots

Plot	pH CaCl2	pH KCl	C %	N %	C/N	K Ekv	P Ekv	S %
Zavodnje (polluted)	3.90	3.82	6.6	0.32	21	0.18	Traces	0.073
Mislinja (unpolluted)	3.66	3.52	14.4	0.62	23	0.19	Traces	0.060

In 1993, during the vegetation season monthly samplings with 270 ml soil cores were taken (at 3 - 5 cores per plot, which equals 21 soil cores per plot). In these all fine root and ectomycorrhizal root tips were counted and the percentages of different types were calculated. Additionaly, soil and mycorrhizae from underneath fungal fruit bodies were taken in order to characterize putative new types of ectomycorrhizae. Classical anatomical methods (AGERER 1987-1995, HAUG & PRITSCH 1992, INGLEBY & al. 1990) and molecular tools (GARDES & BRUNS 1993) were applied for the identification (as described in KRAIGHER & al. 1995). At the time of sampling, fungal species and their occurrence were estimated as a single observation (+), a few (++), frequent

(+++) and massive occurrence (++++). Only the occurrence of ectomycorrhizae from one sampling date (21.10.1993) is presented in figures.

Results and Discussion

In the 21 soil cores from the polluted plot 17 types of ectomycorrhizae were determined on 28443 root tips. The predominant types were *Paxillus involutus, Xerocomus badius, Picerhiza parallela & Piceirhiza inflata.* On the less polluted plot 24 different types were determined on 38502 root tips in equal volume and number of soil cores. The most frequent and constantly present types were *Hydnum rufescens, Amphinema byssoides, Piceirhiza oleiferans, Cenococcum geophilum & Lactarius lignyotus.* All types were briefly described, while *Lactarius lignyotus x P. abies* (KRAIGHER & al. 1995) and *Hydnum rufescens x P. abies* (AGERER & al. in prep.) have been comprehensively described by anatomical and molecular methods.

On the sampling date 21.10.1993 the most frequent types on the polluted plot were determined as *Paxillus involutus, Piceirhiza inflata, Xerocomus badius* and *Piceirhiza parallela* (Fig. 1a). The two identified types have been previously reported to occur on heavily polluted plots in Central Europe (TURNAU & al. 1993). On the same sampling date, the predominant mycorrhizal fruitbodies belonged to *Paxillus involutus (+++), Amanita gemmata (+++), Thelephora terrestris (+++), Xerocomus badius (++), Tricholoma saponaceum (++), Lactarius mitissimus (++)* and *Laccaria amethystina (++).* The presence or absence of fruitbodies could not be correlated with the occurrence of different types of ectomycorrhizae, as reported previously (i.e. SHAW & al. 1992). The predominance of fruitbodies did not linearily follow the abundance in ectomycorrhizal types, i.e. in mycelium, which is present in the soils.

At the same sampling date, the most frequent types on the unpolluted plot were *Hydnum rufescens, Amphinema byssoides, Piceirhiza oleiferans* and *Cenococcum geophilum* (Fig. 1b). We would like to emphasize the high and constant frequency of *Hydnum rufescens* ectomycorrhiza throughout the vegetation season, as hydnoid fungi are known to be among the first fungal species to disappear under the influences of pollution (ARNOLDS 1991). On this sampling date the most frequent mycorrhizal fruitbodies on this plot were *Lactarius lignyotus (+++), Lactarius mitissimus (+++), Laccaria amethystina (+++), Russula ochroleuca (+++), Hygrophorus olivaceoalbus, Cortinarius camphoratus, Cortinarius varius, Cortinarius malachius, Cantharellus tubaeformis, Laccaria laccata, Paxillus involutus, Xerocomus badius, Lactarius subdulcis* (all ++). *Hydnum rufescens* fruitbody was only noticed once on the plot, while a few more have occurred in the vicinity of the plot. Neither *Amphinema byssoides* nor any other corticiaceous fungal species was noticed at all and yet its 'pirate' hyphae and the type itself were rather frequent formers or companions of ectomycorrhiza. A few fruitbodies of *Paxillus involutus* were found in the October sampling in

Mislinja. However, on this sampling date, no ectomycorrhizae of the type were noticed. Therefore, for this plot, it was even more apparent that no clear correlations were possible between fruitbody and ectomycorrhizal type occurrence.

Conclusions

From comparison of the occurrence of ectomycorrhizae on the two forest research plots and fungal fruitbody we derive the following conclusions:

1) Pollution in connection to other environmental factors can influence the distribution of mycorrhizal fungi, whereby several species can disappear while others can profit from the new less competitive stage in the stressed forest ecosystem;

2) Mycobioindication through mycocoenoses is a possible method to apply in the bioindication of forest sites if mycorrhizae are used (as suggested by FELLNER 1989 and ARNOLDS 1988 and 1991);

3) From comparisons of fruitbody occurrence and types of ectomycorrhizae, the latter seems to be superior for establishing the presence of a particular fungus on a forest site, as: i) ectomycorrhizae can be present in forest soils throughout the year and also in different vegetation seasons, while the presence of fruitbodies is restricted to a certain period of the year and their presence can vary from year to year due to general climatic conditions; ii) a great number of ectomycorrhizal fungi belong to Deuteromycotina, which are difficult to identify, or to corticiaceous fungi, whose sporocarps are difficult to observe, as well as to hypogeus fungi, whose sporocarps need special sampling procedures as they are neatly hidden below ground; iii) in several cases on our plots, the identity of the fungal partner of ectomycorrhizae was not known, but ectomycorrhizae seemed to be restricted either to the polluted plot (such as *Piceirhiza inflata* and *Piceirhiza terraphila*) or to the unpolluted plot (such as *Piceirhiza oleiferans* and *Piceirhiza harenosa*);

4) In Slovenia, mycobioindication through selective sensitive or unsensitive fungal species seems to be the choice method for the application of this approach, whereby we suppose that the sensitive species might be *Hydnum rufescens* and the nonsensitive might be *Paxillus involutus*; however, further screening of comparable forest sites influenced differently by pollution is required to select the pollution-sensitive and unsensitive species.

Acknowledgements

We would like to thank A. PILTAVER for fungal fruitbodies identification and Jana Janša for technical assistance. The study was financed by the Ministery for Science and Technology of the Republic of Slovenia and was part of the TEMPUS JEP 04667-93/2 'Bioindication of Forest Site Pollution - Development of Methodology and Training'.

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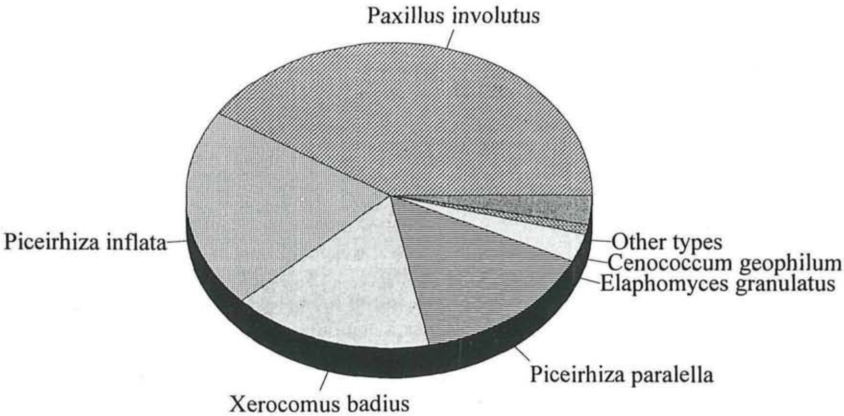

Fig. 1a. Ectomycorrhizal types which occured in soil cores from Zavodnje (the polluted plot) on 21.10.1993.

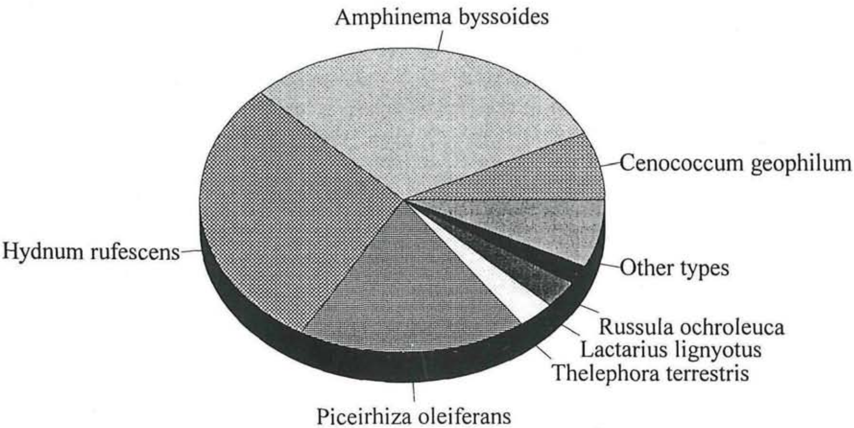

Fig. 1b. Ectomycorrhizal types which occured in soil cores from Mislinja (the unpolluted plot) on 21.10.1993.