4.4 Species composition and diversity of fungi in synanthropic biological soil crusts

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INTRODUCTION

In this chapter we present results of a two-year study dealing with the effect of mechanical disturbance on species composition and diversity of fungi in biological soil crusts on two different localities, where a previous preliminary study of microfungal communities had been made, i.e., on the abandoned ore-washery sedimentation basin at Chvaletice and on an unforested area near the former military airport near Ralsko.

MATERIALS AND METHODS

Localities and sampling of biological soil crusts: Sampling was made in November 2005 and November 2006 on Chvaletice and Ralsko localities (see Colour plates, Figs. 4.1, 4.2). For the characteristics of the localities, see above in the Chapter 4. For the mycological analysis, the same material of biological soil crusts was used as for studying of algae and cyanobacteria. For the sampling pattern see Chapter 4. At Chvaletice and Ralsko, soil crust samples from four and three square plots were taken, respectively. One half of the samples originated from undisturbed nonseeded subplots and one half from disturbed nonseeded subplots. In total, 16 samples of crusts from Chvaletice locality and 12 samples from Ralsko were processed.

Cultivation and identification: Microscopic fungi in biological soil crusts were studied by the cultivation methods described in the Chapter 2.4. For isolation of fungi, three agar media were used: soil agar with rose Bengal and glucose (SEGA), wort agar (WA), and Sabouraud's agar (SAB), all with streptomycin (0.1 g/l) (Fassatiová 1986). Thus, one sample of biological soil crust was inoculated on 12 isolation Petri dishes.

Identification of soil micromycetes was made according to the literature cited in the Chapter 2.4 and according to Ellis (1971), Cannon & Hawksworth (1982), Ramírez (1982), and Arx et al. (1986). Several fungal strains were deposited at Culture Collection of Fungi (CCF), Department of Botany, Charles University, Prague, Czech Republic (see in Tab. 4.4.1).

Statistical methods: Detrended Correspondence Analysis (DCA) and Canonical Correspondence Analysis (CCA) were performed using Canoco 4.5 (Ter Braak & Šmilauer 1998) to ordinate localities based on their microfungal composition.

RESULTS AND DISCUSSION

In Tables 4.4.1 and 4.4.2, microfungi isolated during 2005–06 from disturbed and nondisturbed biological soil crusts on localities Chvaletice and Ralsko are listed. Altogether 46 and 31 taxa (species, forms, and undetermined isolates) of microscopic fungi were recorded from Chvaletice and Ralsko, respectively. The higher figure of microfungal species isolated at Chvaletice compared to Ralsko is probably due the higher number of samples processed (16 versus 12 samples). At Chvaletice, a survey of diversity of microfungi occurring on industrial deposit was made several years ago (Kubátová et al. 2002). During this study, 94 fungal species was recorded, based on 52 samples processed. The higher number of detected microfungi is again obviously due to the higher number of samples processed during the study published in 2002.

As concerns nondisturbed and disturbed subplots at Chvaletice and Ralsko (see Tables 4.4.1 and 4.4.2), overall species richness was similar (35 and 36 fungal taxa in Chvaletice, respectively, and 26 and 25 fungal taxa in Ralsko, respectively). Species richness on individual subplots was in the range of 4–16 fungi recorded.

The majority of the fungi isolated at Chvaletice are anamorphs of Ascomycota (38 taxa, 86%); six taxa (14%) belong to Zygomycota. The most frequent genus was *Penicillium* (10 species). At Ralsko anamorphs of Ascomycota were also prevailing (26 taxa, 90%); three taxa (10%) belonged to Zygomycota. The *Penicillium* again was the richest genus (12 taxa). Similar terms as concerns ascomycetes and zygomycetes we found on other localities studied (see Chapter 2.4 and 3.6). They are similar to data cited in studies dealing with soil microfungi and using conventional isolation methods.

The most frequent fungi of biological soil crusts at Chvaletice were *Trichoderma* spp., *Trichoderma virens*, sterile dark mycelia, *Penicillium janthinellum* (Fig. 4.4.1 a), *P. crustosum*, *P. pulvillorum* (Fig. 4.4.1 b), *Polyscytalum* sp. (Fig. 4.4.1 c), *Penicillium simplicissimum*, and *P.* cf. *coalescens* (Fig. 4.4.1 e). A spectrum of these dominant fungi share four species (*Penicillium janthinellum*, *P. simplicissimum*, *Trichoderma* spp., and *T. virens*) with dominants found in substrate of this abandoned ore-washery basin several years ago (Kubátová et al. 2002). We consider these fungi as highly tolerant to stress factors on the studied localities. Among

Table 4.4.1 Chvaletice (**C1–C4**) – a list of microscopic fungi isolated from disturbed (**D**) and nondisturbed (**N**) biological soil crusts in 2005 and 2006.

Microscopic fungus	C1N		C2N		C3N		C4N		CN		C1D		C2D		C3D		C4D		CD	
	05	06	05	06	05	06	05	06	05	06	05	06	05	06	05	06	05	06	05	06
Acremonium berkeleyanum	2								+		2								+	
Acremonium sp.				1				1		+		1								+
Alternaria cf. *alternata*																	1		+	
Arthrinium arundinis							1		+											
Arthrinium phaeospermum ***CCF 3618***			1						+								1		+	
Aspergillus section *Fumigati*												1								+
Aureobasidium pullulans																	1		+	
Bipolaris sorokiniana	1								+											
Cladosporium cladosporioides		1	1	1					+	+		1	1						+	+
Cladosporium herbarum							1		+			1								+
Cladosporium sphaerospermum				1						+										
Epicoccum nigrum											1	1					2		+	+
Fusicladium sp.			1						+											
Lecanicillium psalliotae												1								+
Lecanicillium sp.		1		1		1				+				2						+
Mortierella sp.		1		1		1				+		1								+
Mucor hiemalis f. *hiemalis*	1								+											
Mucor sp.		1								+		1								+
Oidiodendron sp.					2				+				1						+	
Paecilomyces inflatus ***CCF 3617***			1						+											
Paecilomyces lilacinus			2						+						1				+	
Penicillium cf. *coalescens* ***CCF 3864***					2			1	+	+	1				1	2			+	+
Penicillium citreonigrum							1		+											
Penicillium crustosum		1		1		1		2		+	1	1		1		1			+	+
Penicillium chrysogenum			1						+											
Penicillium janthinellum	1		1	1	1	1			+	+	2		1		2	2	1		+	+
Penicillium olsonii														1						+
Penicillium pulvillorum ***CCF 3712***	1				1	1			+	+	1	1			1	1			+	+
Penicillium sacculum																	1		+	
Penicillium simplicissimum ***CCF 3711***		2	1		2				+	+							2		+	
Penicillium spp. (2)		1								+					1				+	
Phoma sp.		2								+		1					1		+	+
Pithomyces chartarum																	1		+	
Polyscytalum sp.		1	2	1	1	1			+	+								1		+
Sphaerodes fimicola							1		+											
Sporormiella sp.												1								+
sterile dark mycelia			1	1	1	1			+	+		1					1	1	+	+
sterile light mycelia				1						+	1								+	
Trichoderma harzianum		2								+		2					1		+	+
Trichoderma virens	1	2		2	1	2	1	2	+	+	2	2	2	2		2		2	+	+
Trichoderma spp. (2)	2	1	2	1	2	2	2	2	+	+	2	1	2	2	2	2	2	2	+	+
Umbelopsis angularis											1								+	
Umbelopsis isabellina			1			1			+	+					1	1	1		+	+
Umbelopsis ramanniana											1								+	
No. of taxa on several subplots	**7**	**12**	**12**	**12**	**9**	**10**	**6**	**5**			**11**	**16**	**5**	**5**	**7**	**7**	**13**	**4**		
Total No. of taxa: 46	**34**								**24**	**22**	**36**								**26**	**23**

Note: **1** = rarely isolated species, **2** = frequent species, CCF = Culture Collection of Fungi, Prague, CZ

Table 4.4.2 Ralsko (**R1–R3**) – a list of microscopic fungi isolated from disturbed (**D**) and nondisturbed biological soil crust (**N**) in 2005 and 2006.

Microscopic fungus	R1N		R2N		R3N		RN		R1D		R2D		R3D		RD	
	05	06	05	06	05	06	05	06	05	06	05	06	05	06	05	06
Acremonium berkeleyanum	1						+									
Acremonium cf. *strictum*										1						+
Acremonium sp.		1						+	1				1		+	
Alternaria alternata													1	1	+	+
Aspergillus clavatus					1		+									
Chaetomium aureum ***CCF 3624***		1		1		1		+	1			2			+	+
Cladosporium cladosporioides			1				+			1						+
Epicoccum nigrum	1		1		1		+									
Mucor spp. (2)	1					1	+	+	1					1	+	+
Penicillium aculeatum ***CCF 3863***	1	1		1	1	2	+	+	2	1	1	2		2	+	+
Penicillium brasilianum			1				+									
Penicillium canescens				1				+				1				+
Penicillium chrysogenum			1				+				1				+	
Penicillium janthinellum ***CCF 3713***	1	2	1	2	1	2	+	+		1		1	1	1	+	+
Penicillium miczynskii ***CCF 3722***										2						+
Penicillium pulvillorum			1				+					2	1		+	+
Penicillium purpurogenum						1		+								
Penicillium purpurogenum var. *rubrisclerotium*									1						+	
Penicillium smithii	1	1	1	2		1	+	+	2				1	1	+	+
Penicillium spinulosum				1		1		+		1		1		1		+
Penicillium spp.	1	1	2	1	1		+	+		1	1	1	1		+	+
Pochonia bulbilosa				1				+	1					1	+	+
Polyscytalum sp.			1				+									
Sphaerodes fimicola										1				1		+
sterile dark mycelia		1	1	1			+	+	1		1				+	
Trichoderma viride		1						+		1			1		+	+
Trichoderma spp. (2)	2	1	2	2	2	2	+	+	1	2	2	1	2	2	+	+
Umbelopsis angularis	2	2	2	2	1	2	+	+	1	2	1	2	1	2	+	+
Umbelopsis isabellina	1	2		2	1	2	+	+	2	2	1	2		2	+	+
No. of taxa on several subplots	**10**	**11**	**12**	**12**	**8**	**10**			**11**	**12**	**7**	**10**	**9**	**11**		
Total No. of taxa: 31	**26**						**19**	**18**	**25**						**19**	**21**

Note: **1** = rarely isolated species, **2** = frequent species, CCF = Culture Collection of Fungi, Prague, CZ

the above cited frequent fungi, *P. crustosum*, *Polyscytalum* sp., and *P.* cf. *coalescens* fall into unexpected findings. *Penicillium crustosum* is a typical food-borne species; however, though laboratory contamination was excluded, it was found in both studied years. *Polyscytalum* sp., and *P.* cf. *coalescens* belong to a somewhat rare soil and litter fungi. The number of fungi recorded at Chvaletice only sporadically was lower then on other localities studied (see Chapt. 2.4 and 3.6); the majority of them belong to micromycetes known from soil and plant remains (Domsch et al. 1993). Noteworthy among them are *Aspergillus* section *Fumigati* and *Cladosporium* sp. *Isolate* of *Aspergillus* section *Fumigati* probably presents one of the most newly described species in the section *Fumigati* (Hong et al. 2005), not yet known from the Czech Republic. Isolate of *Cladosporium* sp. is obviously a lesser known species of this common genus. Both isolates will be analyzed by molecular methods.

The dominant microfungi of biological soil crusts at Ralsko appeared to be *Trichoderma* spp., *Umbelopsis angularis*, *U. isabellina*, *Penicillium aculeatum*, *P. janthinellum*, *P. smithii* (Fig. 4.4.1 d), *Penicillium* spp., and *Chaetomium aureum*. Compared with the Chvaletice locality, it is a different spectrum of fungi, sharing only one species, *Penicillium janthinellum*. The high occurrence of this species is considered as evidence of some stressing factors at this site. Other species like *Umbelopsis angularis*, *U. isabellina* or *P. smithii* were found often on natural localities (e.g., Šumava Mts., Kubátová et al. 1998). On the other hand, *P. aculeatum* is rather rare species (Pitt 1979) as well as *Chaetomium aureum*. This fungus is known from natural substrates. How-

ever, it was isolated also from soils and litter polluted with radionuclides near Chernobyl, Ukraine and is considered a bioindicator of radionuclide polluted soils (Zhdanova et al. 1995, 2001, 2005). In our country it was found for the first time (see Kubátová 2006). Looking at the microfungal community at Ralsko, we can consider this locality similar to natural localites, influenced some stress factors however.

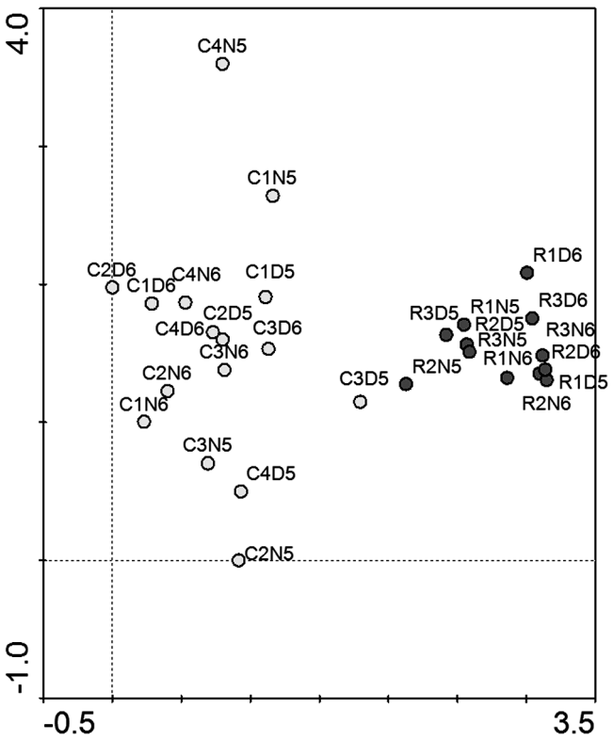

Fig. 4.4.2 Testing of differences in fungal species richness and composition of biological soil crusts on disturbed and nondisturbed subplots at Chvaletice and Ralsko during two years. DCA ordination diagram showing the position of samples in the range of the first two ordination axes (**C1–C4** = Chvaletice, **R1–R3** = Ralsko, **D** = disturbed subplots, **N** = nondisturbed subplots, **5** = 2005, **6** = 2006).

Species composition of fungal communities recorded at Chvaletice and Ralsko is generally very different. From 16 sharing species, only one species is dominant for both sites (*Penicillium janthinellum*). The CCA analysis of species richness and composition significantly differentiated both sites (p-values 0.0005). For comparison of species richness and composition using DCA, see diagram (Fig. 4.4.2). The CCA analysis testing difference in species composition during two years of sampling (2005 and 2006) significiantly confirmed great differences in species composition between samples processed in the different years, although the p-values were higher than in the case of locality effect testing (p-values 0,001). The effect of disturbance on species composition tested by paired T-test proved to be not significant (0.72).

CONCLUSIONS

Microfungi isolated from biological soil crusts of the abandoned ore-washery basin at Chvaletice and from an unforested site near former military airport Ralsko, belong mostly to the soil and litter fungi similarly as in previous studies. On both localities, only small differences were found in fungal species richness recorded on disturbed and nondisturbed subplots. Species composition of fungal communities recorded at Chvaletice and Ralsko proved to be very different. On both localities, more dominant species of fungi were recorded than on localities in previous studies. Among the dominant fungi of biological soil crusts at Chvaletice are fungi considered as stress tolerant. The fungal species composition of biological soil crusts in Ralsko is closer to that of natural forested habitats.

Significant differences in fungal species composition were proved among biological soil crusts sampled in different years. In Ralsko, the interesting radionuclide tolerant fungus *Chaetomium aureum* was found. It is the first report of this fungus for the Czech Republic.