4.2 Species composition and diversity of algae in synanthropic biological soil crust

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INTRODUCTION

This chapter provides comparison between the algal flora of ore-sedimentation sedimentation basin Chvaletice and natural forest-free area near Ralsko village. Beyond the general comparison of algal floras, we refer detailed descriptions of several taxa found in the investigated localities.

MATERIALS AND METHODS

In each site, samples from the 0–3cm layer were taken randomly from several physiognomically uniform square areas (eight in Chvaletice – C1A, C1D, C2C, C2D, C3B, C3C, C4A, C4D and six in Ralsko – R1A, R1C, R2B, R2D, R3B, R3C). All samples were collected twice, in 2005 and 2006. The samples were placed into sterile bags and transported to the laboratory for analysis. The composite samples were crushed and mixed to produce a homogenouos sample. A 1-g aliquot was removed and added to 50 ml of distilled water. The soil suspension was mixed by a magnetic mixer for 15 minutes. Aliquots of 0.5 or 1 ml were spread in duplicate on agar solidified BBM and WC medium (Andersen 2005). Cultures were sealed with parafilm and incubated at 20–25 °C under daylight conditions (the plates were placed beside a north facing window) until good growth had been obtained (3–6 weeks). Algal microcolonies were examined directly from agarized plates using an Olympus BX 51 microscope with Nomarski DIC optics and photographed using Olympus Camedia digital camera C-5050 Zoom. Standard cytological stains (Lugol's solution, methylene blue, acetocarmine, chloraliode solution) were used for visualisation of pyrenoid, cell wall structures or mucilage. For detailed investigation of some strains, the algal colonies were transferred to agarized BBM culture tubes and then cultivated at 18 °C, under an illumination of 20–30 μmol $m^{-2} \cdot s^{-1}$ and 16:8 h light-dark cycle. Identification was made on the basis of life history and morphological criteria using standard authoritative references (Fott & Nováková 1969; Ettl 1978; Komárek & Fott 1983; Punčochářová 1992; Ettl & Gärtner 1995; Hindák 1996; Lokhorst 1996; Andreeva 1998). The quantities of algae on Petri dishes were evaluated as belonging to one of three classes: 1 – single algal colony found, 2 – rare species with several colonies on the dishes, 3 – dominant species in the sample. Following statistical methods were used to detect patterns in the data. Dice floristic similarities between selected sampling sites were counted in statistical program PAST 1.74 (Hammer et al. 2001). 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 algal composition. In CCA, 2000 permutations were performed to test the given hypotheses.

RESULTS AND DISCUSSION

General conclusions

A total of 34 algal species representing 25 genera were recovered during our investigation of several sampling areas in Chvaletice and Ralsko (Table 4.2.1). Six widespread taxa (coccal green algae *Watanabea* sp., *Pseudococcomyxa simplex* and *Radiococcus* sp.; and filamentous algae *Geminella terricola*, *Klebsormidium flaccidum* and *Stichococcus bacillaris*) were found in more than ten of 28 samples. We implied from this that green algae represented the most dominant algal group. In both localities, the members of Chlorophyta comprised 65% majority of all determined taxa (Fig. 4.2.1). Surprisingly, no diatoms and scarcely any cyanobacteria were determined during the investigation. That demonstrated a distinct difference when compared to the algal composition of biological soil crusts on natural substrata (see Chapter 2.2).

Comparing the species richness of both localities, Ralsko encompassed obviously more taxa than Chvaletice, even if fewer sampling sites were investigated there (Fig. 4.2.2). In spite of a similar proportion of algal groups determined in these localities, their species composition significantly differed. The CCA analysis significantly differentiated both localities on the basis of algal flora, whether it was tested separately for each year or for both years together (p-values invariably 0.0005). The distinct separation of Chvaletice and Ralsko samples is also well illustrated in the DCA ordination diagram (Fig. 4.2.3). It all corresponds with different dominant algal species for both localities. While *Radiococcus* sp., *Watanabea* sp. and *Pseudococcomyxa simplex* dominated in Chvaletice, Ralsko was characterized by the dominance of *Ulothrix tenerrima* and *Klebsormidium flaccidum* (see Table 4.2.1).

Table 4.2.1 Algal distribution in all investigated sampling sites (for explanation of the abbreviations see Materials and Methods).

Year of sampling	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006
Sampling site	C1A		C1D		C2C		C2D		C3B		C3C		C4A		C4D		R1A		R1C		R2B		R2D		R3B		R3C	
Cyanobacteria																												
Leptolyngbya sp.			2																									
Nostoc sp.	1		1																									
Xanthophyceae																												
Botrydiopsis cf. *arhiza* A. Borzi						2																						
Heterococcus sp.																												1
Eustigmatophyceae																												
Eustigmatos magnus (J. B. Petersen) Hibberd																											1	
Eustigmatos vischeri Hibberd																					3	1		1		2		
Chlorophyceae																												
Bracteacoccus sp.	2	2	2	1															1								1	
Desmococcus sp.																	3		1			2						1
Diplosphaera chodatii Bialosuknia em. Vischer	1	2		3		2		1		1		1											2	2				
Mychonastes homosphaera (Skuja) Kalina et Puncocharova																			1									1
Radiococcus sp.	3	2	3	3		1	2	3	2					3	3	2												
Trebouxiophyceae																												
Asterochloris sp.							1	1							1	1												
Chlorella mirabilis Andreeva					2	1																						
Chlorella cf. *luteoviridis* Chodat																						2						
Chlorella cf. *trebouxioides* Puncocharova																			1	1			2	2				
Chlorella sp.																												2
Choricystis chodatii																												1
Coenocystis cf. *oleifera* (Broady) Hindák	1	2	3	1					1	3		3							1				1					
Leptosira erumpens					1	1	1						2		2								1	1				
Muriella zofingiensis (Dönz) Hindák																							1					
Myrmecia incisa Reisigl																		3										
Myrmecia sp.	2	2	2			3	1	2					2		2	2			1									
Pseudococcomyxa simplex (Mainx) Fott	1	1		1	2	3	2		3	3		1	1	2		1		2			1							
Stichococcus allas Reisigl																								1				
Stichococcus bacillaris Nägeli	2	3	3	2	3	2			1	3	2	3	1	1		1		1				2		2				2
Watanabea sp.	3	3		3	3	3	3	3	2	1	1	2	3	3	3	3		1				1	1					
Ulvophyceae																												
Kentrosphaera gibberosa Vodenicarov et Benderliev																			2	2					2	1		
Ulothrix tenerrima Kützing																	2	2			2	3	2	2				
Klebsormidiophyceae																												
Geminella terricola Boye Petersen	1	1	1	1	3	3	2	1	2						2	2				3			1					
Klebsormidium dissectum (Gay) Ettl & Gärtner			3																									
Klebsormidium flaccidum (Kützing) Silva, Mattox et Blackwell	1	1		1					1								1	2	2		2	3	2	2	2	3	3	
Klebsormidium fluitans (Gay) Lokhorst																		3										
Klebsormidium mucosum (Boye Petersen) Lokhorst																					1	1						
Zygnematophyceae																												
Mesotaenium sp.																		1										

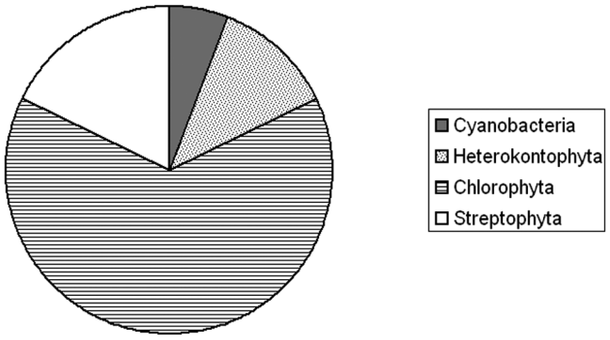

Fig. 4.2.1 Proportional occurrence of four algal groups, determined in all investigated localities.

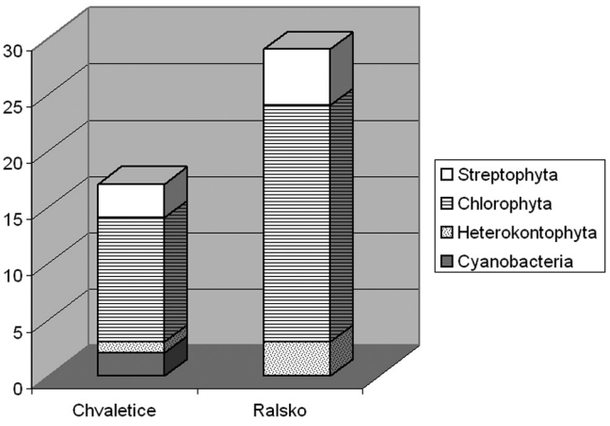

Fig. 4.2.2 Species richness expressed as the number of taxa found in each locality. Assignment of taxa to the four algal groups is displayed.

Since the same sampling sites were investigated in 2005 and 2006, the change of species composition during this period can be investigated. DCA ordination plot in Fig. 4.2.4 illustrates the pattern of these changes, connecting graphically the same sampling sites during two years. The diagram obviously indicates greater difference in species composition at Ralsko. The same result is obtained, comparing the average Dice coefficients of the species composition similarities counted for each couple of the samples from the same sampling site, but differing by the date (Chvaletice – 0.713, Ralsko – 0.398). Hence, Ralsko is characterized by a quite large transformation of species composition, which took place between 2005 and 2006. However, we did not determine any common pattern in the year-on-year shift in species composition, either in Ralsko (CCA analysis, p-value 0.83), nor in Chvaletice (CCA analysis, p-value 0.22). Therefore, even if some differences on species composition were observed in both localities, these have no common pattern and are caused rather by a unique, stochastic change within each sampling site.

Finally, the DCA ordination was performed to illustrate the correlation of measured physico-chemicals characteristics as well as their correlation with the observed differences in species composition of investigated samples (Fig. 4.2.5). Differences in the algal composition of Chvaletice and Ralsko well correspond to the diverse values of conductivity, phosphorus concentration (both higher in Chvaletice) and nitrogen concentration (higher in Ralsko). Further, the higher rates of nitrification, nitrate ammonification and chlorophyll a concentration advert to the accelerated biological activity in Ralsko.

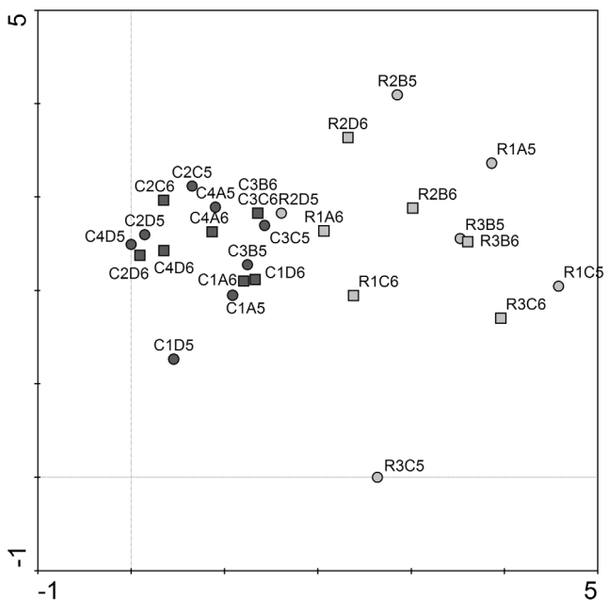

Fig. 4.2.3 DCA ordination diagram showing the position of samples in the range of the first two ordination axes (dark – Chvaletice, bright – Ralsko; circle – 2005, square – 2006).

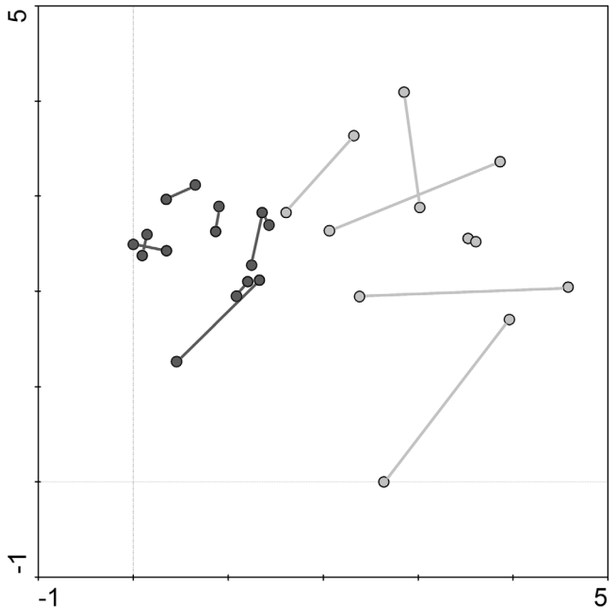

Fig. 4.2.4 DCA ordination diagram showing the position of samples in the range of the first two ordination axes (dark – Chvaletice, bright – Ralsko). The same sites sampled in 2005 and 2006 are connected to illustrate the year-on-year changes in the species composition.

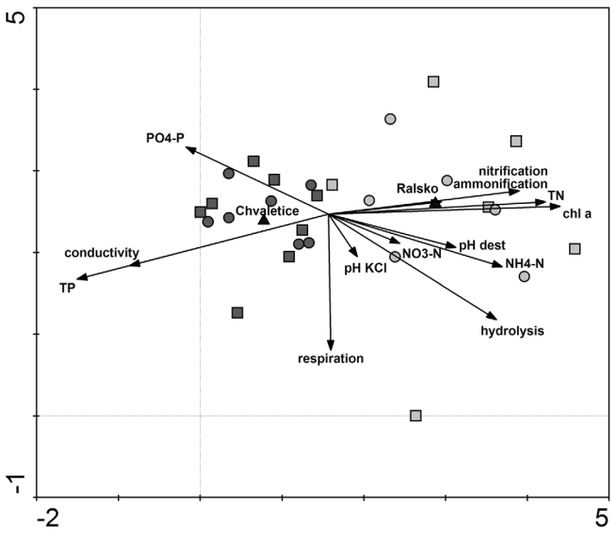

Fig. 4.2.5 DCA ordination diagram showing the position of samples and environmental characteristics in the range of the first two ordination axes (dark – Chvaletice, bright – Ralsko; circle – 2005, square – 2006).

Floristics

The following pages provide detailed descriptions of several interesting taxa found in the investigated localities (see Colour plates, Figs. 4.2.6, 4.2.7).

***Myrmecia* sp.** (Figs. 4.2.7 a–c)

A very remarkable coccal green alga was determined in several sampling sites in Chvaletice. It was characterized by always spherical cells with smooth cell walls. The young cells contained one parietal incised chloroplast. In mature cells, the chloroplast divided into several flat parts, lying parietally beneath the cell wall. Occasionally, the flat chloroplasts forming the second layer below the first one could locally appear. The cells were uninucleate, with well visible nucleus and nucleolus. Almost always, a large distinct vacuole was situated near the cell centre and nucleus. The cells reached 15(–19) μm in diameter. The asexual reproduction took place by means of 8–32 spherical autospores. It is noteworthy that the cells were determined only from the agar plates with WC cultivation medium.

The taxonomic position of this organism is unclear. Cell dimensions and parietal position of chloroplast corresponds to the definition of *Myrmecia* (Ettl & Gärtner 1995). However, *Myrmecia* contains only one chloroplast, though distinctively incised in some species. The same could be stated to distinguish it from genus *Lobosphaera*. Possession of several chloroplasts per cell is typical for *Muriella* species. These, however, generally do not reach the dimensions of the observed alga. Moreover, in *Muriella* the distribution of chloroplasts is simpler than observed in investigated alga, constituting only one chloroplast layer beneath the cell wall. The genus *Bracteacoccus*, typical by large cells possessing many chloroplasts, is characterized by multinuclear cells. However, we always observed only single nucleus per cell.

Accordingly, we are not able to determine either the specific epithet or the generic name of observed alga. However, the characteristic morphology incongruent with any described algal species refers to the discovery of a still undescribed green algal genus.

***Watanabea* sp.** (Figs. 4.2.7 f–i)

In all but one sampling site in Chvaletice, we determined the distinct green coccal alga *Watanabea* sp. In 11 samples, it forms a dominant component of the algal community. The cells were characterized by spherical to oval shape; with dimensions varying in the range of 6–14 × 5–12 μm. Cell wall was smooth. Cells were uninucleate, possessing single chloroplast with distinct pyrenoid. Chloroplast was parietal, initially discoid, sometimes becoming band-shaped. At maturity, chloroplast developed into more complex stage, either parietal with the incisions and long sub-plasmatic chloroplast lobes or axial with short irregular lobes spreading the cell periphery. Asexual reproduction took place by autospores, two to four spherical spores or 8–32 ellipsoidal to cylindrical spores produced per sporangium.

The above-characterized organism corresponds morphologically well with the definition of *Watanabea*, the relatively recently described green algal genus of Trebouxiophyceae (Hanagata et al. 1998). In particular, the pattern of asexual reproduction is identical for both organisms. In *Watanabea*, cells are similarly reproduced by means of two types of aplanospores, the spherical S-form and elliptical E-form ones. However, the only described species of *Watanabea*, *W. reniformis*, is characterized by the chloroplast without pyrenoid. Since we observed a distinct pyrenoid within the chloroplast, investigated alga probably represent a new species of the genus *Watanabea*. However, a confident taxonomic position of this strain could be solved only with the appropriate aid of the molecular biology techniques.

Kentrosphaera gibberosa Vodenicarov et Benderliev

Kentrosphaera gibberosa has been determined in two sampling sites in Ralsko, in both years of study. Large cells of linear, lanceolate, ovoid, globular or irregular shape contained one massive, axile chloroplast, which contained several pyrenoids. Pyrenoids were arranged in a row or equally distributed around the nucleus. Cells were always uninucleate, with the nucleus lying in the central area of the cell. Cell wall was thick, with characteristic lamellate thickenings of the cell wall. Asexual reproduction took place by numerous spherical autospores. This species is considered to be relatively rare, occurring sporadically in aquatic or terrestrial environments (Punčochářová 1992).

According to Wujek & Thompson (2005), *K. gibberosa* is not a valid name yet, since *Kentrosphaera* should be considered as a synonym of *Scotinosphaera*, a green alga described two years earlier (Klebs 1881). Hence, a new combination *Scotinosphaera gibberosa* should be used according to their opinion. However, we do not agree with the presented synonymisation of *Kentrosphaera* and *Scotinosphaera*, mainly because of some distinct incongruencies in the description of these two genera. For example, *Scotinosphaera* was described as having a parietal chromatophor forming a layer

round the whole cell, containing many pyrenoids (Klebs 1881; Blackman & Tansley 1902). In *Kentrosphaera*, however, a single axial chloroplast with few to several pyrenoids was described. It is possible that Klebs (1881) coincidentally described "*Kentrosphaera*" in the stage of zoosporogenesis. This stage is characteristic in possessing many parietal chloroplasts with single pyrenoids. However, we cannot be sure about it and the above-mentioned synonymisation is therefore questionable. Instead, we rather use the generic concept of *Kentrosphaera* sensu Punčochářová (1992).

Ulothrix tenerrima Kützing (Fig. 4.2.7j)

Long filaments of *Ulothrix tenerrima* were found in three sampling sites in Ralsko. In 2006, they even formed a dominant compound of algal flora in the sampling site R3B. The filaments consisted of cylindrical cells with ring-shaped chloroplast and one indistinct pyrenoid. The cell wall was rather thin locally, however, with dominant H-shaped cell wall thickenings. Cells were 7–8 μm wide and 2–7 μm long.

This predominantly freshwater species was rarely recorded from various terrestrial biotopes in Australia, England and USA (Cameron 1964; Ettl & Gärtner 1995). Interestingly, it was determined as a natural compound of desert biological soil crust communities at Camp Floyd State Park in Utah (Johansen et al. 1984).

CONCLUSIONS

We compared algal communities between the biological soil crusts in Chvaletice ore-sedimentation basin and the former military airport near Ralsko. With 27 determined taxa, Ralsko was detected to be more species-rich than Chvaletice (only 17 species determined). Algal composition significantly differed between the localities, but some similarities could be found. In both localities, the members of Chlorophyta comprised a majority of all determined taxa, with no diatoms and scarcely any cyanobacteria found. In Ralsko, we noted large differences of species compositions, comparing the samples in 2005 and 2006. However, these differences do not show any common pattern on a scale of the whole locality; and are caused rather by a unique, stochastic change within each sampling site.