

### 3. Life Sciences (Biosciences)

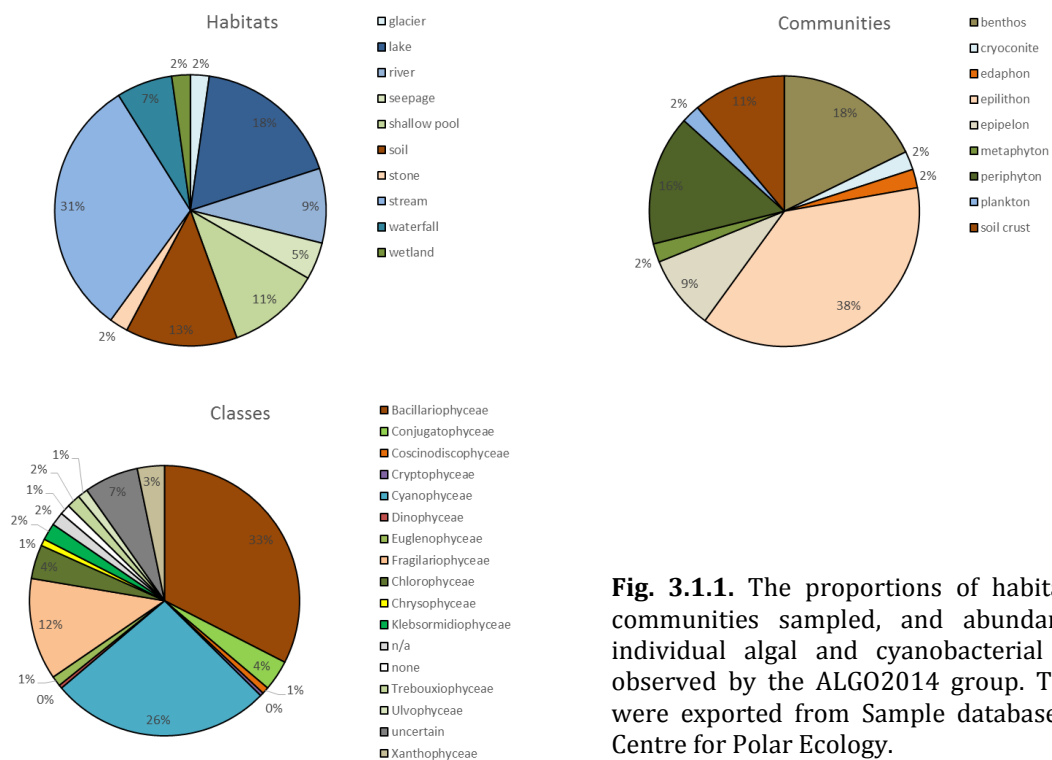
#### 3.1. Microbiology and Phycology

Instructors: *Josef Elster, Jana Kvéderová & Otakar Strunecký*

Students: *Terezie Englová, Klára Hajšmanová, David Ryšánek & Luděk Sehnal*

The long-term aim of the microbiology/phyecology group is to characterize the microbial diversity of algae and cyanobacteria in various freshwater and aero-terrestrial biotopes (streams, pools and lakes, seepages, soil surface, wet rocks, snow, snow cryoconites). We focus not only on taxonomical diversity, but also on diversity in ecology and physiology.

In 2014, total of 43 samples were collected at 23 different localities during the course and we found 121 species/genera identified at species or genus levels. The proportions of sampled habitats, communities and abundance of individual classes of algae and cyanobacteria are summarized in Fig. 3.1.1.

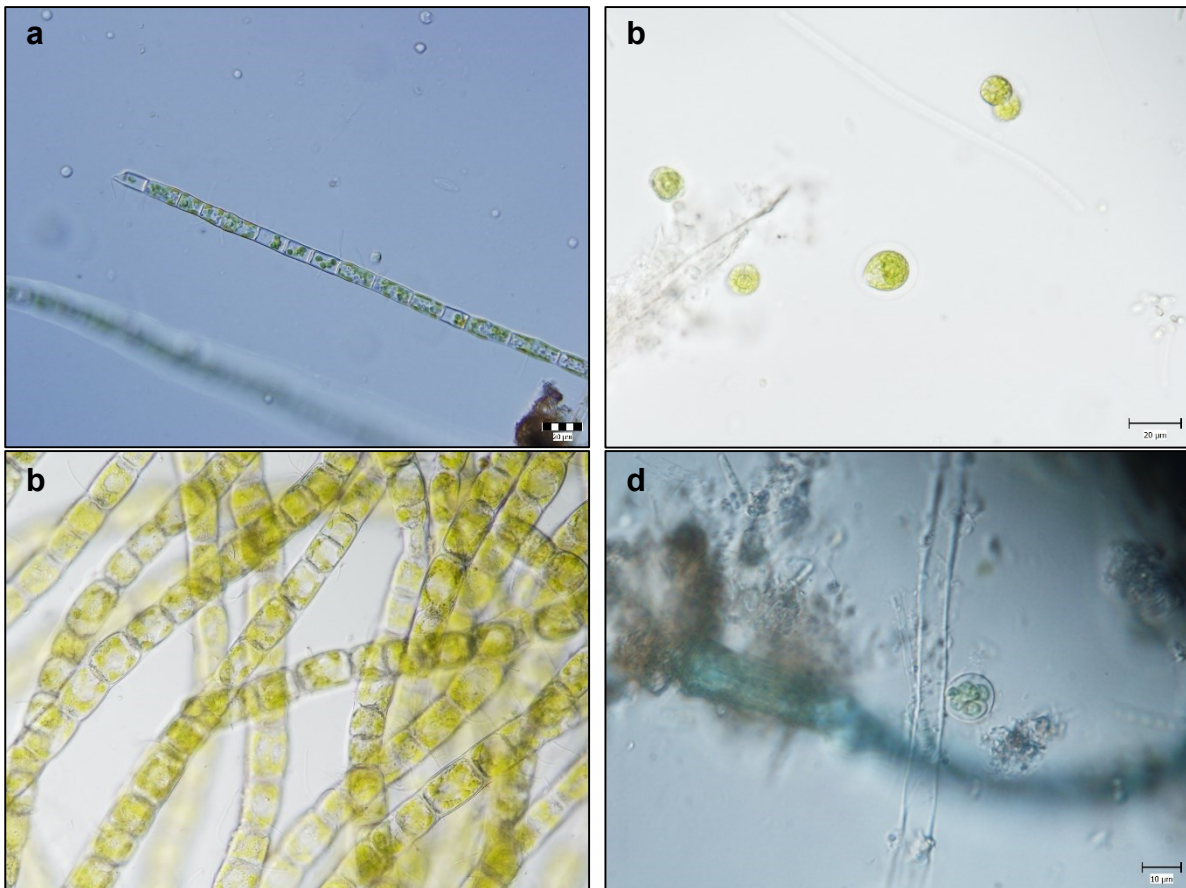


**Fig. 3.1.1.** The proportions of habitats and communities sampled, and abundances of individual algal and cyanobacterial classes observed by the ALGO2014 group. The data were exported from Sample database of the Centre for Polar Ecology.

#### Diversity of phototrophic microorganisms

In total we found 121 genera of organisms, including 30 genera of cyanobacteria, 19 genera/species of green algae *sensu lato* (Chlorophyta and Streptophyta), and 50 genera/species of diatoms.

Majority of taxons observed, like *Nostoc* sp., *Hydrurus foetodus* or *Hannaea arcus*, was the same as in previous years. Contrary, we observed yellow-green alga genus *Tribonema* more frequently (Fig. 3.1.2a.), especially in streams where green filamentous algae had dominated. In small pools eutrophized by bird excrements, the algal community was dominated by *Euglena* sp. and a green flagellates of genera *Lobomonas* sp. and *Chlamydomonas* sp. (Fig. 3.1.2b.). At pond effluent, the cascade was covered by *Ulothrix* sp. (Fig. 3.1.2c.). In soil crust near bird nests, taxons typical for soil crusts elsewhere were observed, like *Nostoc* sp. or *Gloeocapsa* sp. (Fig. 3.1.2d.)



**Fig. 3.1.2.** Some interesting algal taxa observed during Polar Ecology Course 2014. **(a)** *Tribonema* cf. *vulgare*, **(b)** *Chlamydomonas* sp., **(c)** *Ulothrix* sp., **(d)** *Gloeocapsa* sp.

### Measurements of microbial activity in soils

Klára Hajšmanová focused on diurnal measurement of microbial activity and greenhouse gasses emissions of methane ( $\text{CH}_4$ ) and nitrous oxide ( $\text{N}_2\text{O}$ ) in soils with different vegetation developed in Petuniabukta, Billefjorden.

There were selected three different locations: dry meadows, soil crusts and wet meadows (Fig. 3.1.3). On these three selected locations there were determined greenhouse gasses emission of  $\text{CH}_4$  and  $\text{N}_2\text{O}$  with different types of vegetation. Measurements were made using chambers in a few reps on the variant from 12 to 29 August 2014. At each site of measurement were taken soil samples for laboratory analysis (Fig 3.1.4).

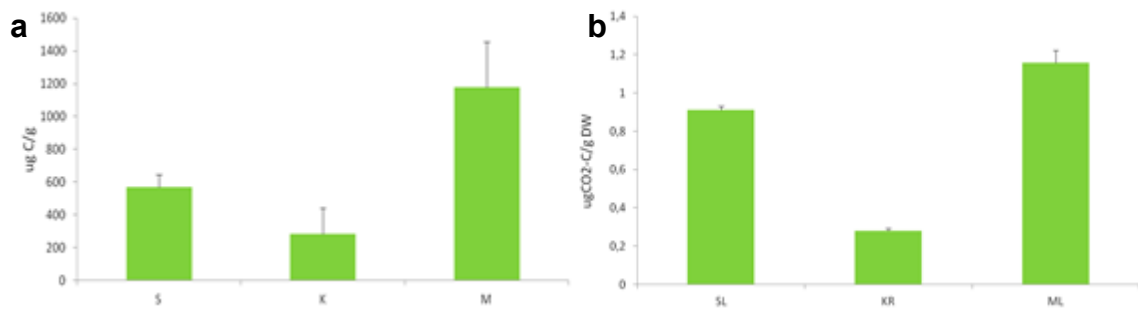


**Fig. 3.1.3.** Field measurement of greenhouse gasses production.



**Fig. 3.1.4. (a)** Air sampling for emissions of methane ( $\text{CH}_4$ ) and nitrous oxide ( $\text{N}_2\text{O}$ ), **(b)** soil sampling.

The results show that with the increasing involvement of vegetation, microbial activity is growing. It is connected with maintaining constant microclimate, large quantities of nutrients and organic matter. Methane emissions further depends on the thawing rate, level of water, temperature and humidity of locations. With the increasing soil temperature, the activity of soil microorganisms increases too and therefore is low to negative values for soil crusts for dry and wet meadows positive emissions. On unfrozen and flooded areas (saturated), methane production prevails over oxidation and there are positive emissions. On unfrozen, drier areas, but not completely dried out there is the decomposition of organic matter, and oxidation of methane emissions prevails over production and there are negative emissions. For unfrozen and slightly flooded soils is production of methane and also oxidation of methane (Fig. 3.1.5.).



**Fig. 3.1.5. (a)** Microbial biomass and **(b)** respiration in different vegetation types. Abbreviations: S – Dry meadows; K – Soil crusts; M – Wet meadows.

### Diurnal cycles of photosynthetic processes

Luděk Sehnal measured diurnal cycles of photosynthetic processes in soil crust and lichen, which were carried out in Petuniabukta, Spitsbergen. For field measurements, a method of induced fluorescence of chlorophyll was used. Measurements of photosynthetic activity were taken as repetitive measurements of effective quantum yield of photosystem II ( $\Phi_{\text{PSII}}$ ). The short-term field measurements were carried out for 10 days in summer 2014.  $\Phi_{\text{PSII}}$  was recorded each 5 minutes as well as microclimatic data (air temperature, air humidity, photosynthetically active radiation - PAR). For measuring of diurnal course of  $\Phi_{\text{PSII}}$  and PAR we used multichannel monitoring fluorometer Moni-PAM 2000 (Heinz Walz, Germany; Fig. 3.1.6.). The fluorometric system was installed in a close vicinity of Petunia station located at Petunia Bay from 5 to 15 August 2014. To determine changes of environmental conditions during the field experiment,



basic microclimatic data were recorded. Temperature and relative air humidity were measured in each five minute by Minikin datalogger (EMS Brno, Czech Republic).

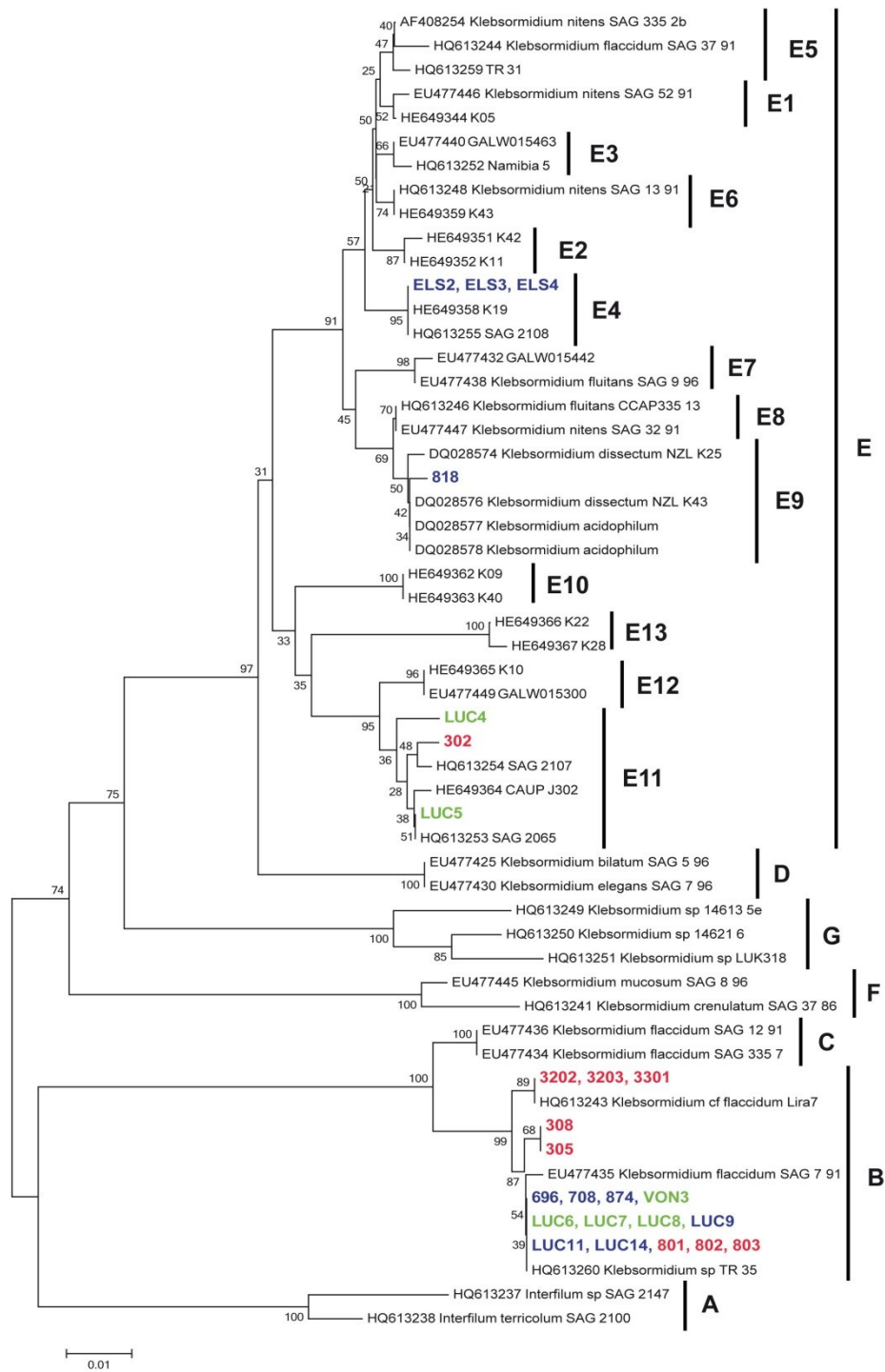
In general, physiological activity of both biological soil crust and lichen showed daily courses. Typically, most of  $\Phi_{PSII}$  values ranged 0.6 – 0.7 in both model organisms. The results have shown that photosynthetic activity was strongly correlated with all observed abiotic factors in both study objects. Particularly important was the relation found between PAR and  $\Phi_{PSII}$  in soil crust. When the soil crust was exposed to high PAR doses of irradiation (about  $2300 \mu\text{mol m}^{-2} \text{s}^{-1}$ ) photoinhibition of primary processes of photosynthesis was observed as  $\Phi_{PSII}$  decrease, while photosynthetic activity of lichen remained at same level. Furthermore, it has been demonstrated increasing that *in situ* photosynthetic activity increased in both soil crust and lichen with a decrease in temperature.



**Fig. 3.1.6.** Fluorometric system Moni-PAM (Walz, Germany).

### Diversity of genus *Klebsormidium* in polar region

In temperate zone the genus *Klebsormidium* is very diverse and has cosmopolitan distribution (Rindi et al. 2011, Ryšánek et al. 2014). Current knowledge showed that strains in this genus have strong ecological preferences (Novis 2006, Škaloud and Rindi 2013, Škaloud et al. 2014), and substrate specificity looks like more important than geographic distribution. On an already obtained strains from arctic and Antarctic from bone proved that same genotype are occurred in both polar regions and also temperate zone on same substrate. So David Ryšánek wanted to collect the same habitats (sandstone, limestone, basalt etc.) like in temperate zone (the Czech Republic) to compare importance of substrate and geographic distribution on diversity of the genus *Klebsormidium*. In Svalbard, David collected 26 samples from sandstone, coal, basalt, soil crust, limestone, orthogenesis, etc. Then on laboratory every samples were maintained on two agar plates slants with modified Bold's Basal Medium (Bischoff and Bold 1963) at  $15^{\circ}\text{C}$  under 24h of light. Algal microcolonies grown up after 6-10 weeks and were isolated into to single strain cultures. In total, 9 strains (3 strains from sandstone, 3 from coal, 2 from basalt, 1 orthogenesis) were used for molecular analysis. The sequences of the *rbcL* gene were obtained by polymerase chain reaction (PCR). It was obtained 4 genotypes from 9 strains. The result showed that the diversity on Svalbard is very low to compare to temperate regions, more often David found mainly genus *Xanthonema* on agar plates, which have very similar morphology and are also more typical for the polar regions (Rybalka et al. 2009). Majority strains from polar are from clade B and strains from clade B had cosmopolitan distribution, they are found arctic, Antarctic and temperate zone. (Fig. 3.1.7.).



**Fig. 3.1.7.** Phylogenetic tree obtained from Bayesian analysis based on *rbcL* dataset, showing the position of investigated *Klebsormidium* strains and their relatives. Red colour show David's strains from Svalbard, green colour show strains from arctic region, and blue colour show strains from Antarctica.

#### References:

- Bischoff H, and Bold HC (1963) Some soil algae from Enchanted Rock and related algal species. Phycological Studies IV. Univ Texas Publ 6318: 1-95.
- Novis PM (2006) Taxonomy of *Klebsormidium* (Klebsormidiales, Charophyceae) in New Zealand streams and the significance of low-pH habitats. Phycologia. 45:293-301.

- Rindi F, Mikhailyuk TI, Sluiman HJ, Friedl T, López-Bautista JM (2011) Phylogenetic relationships in *Interfilum* and *Klebsormidium* (Klebsormidiophyceae, Streptophyta). *Mol Phylogenet Evol.* 58:218-231.
- Rybalka N, Andersen RA, Kostikov I, Mohr KI, Massalski A, Olech M, Friedl T (2009) Testing for endemism, genotypic diversity and species concepts in Antarctic terrestrial microalgae of the Tribonemataceae (Stramenopiles, Xanthophyceae). *Environ Microbiol* 11(3): 554-565.
- Ryšánek D, Hrčková K, Škaloud P (2014) Global ubiquity and local endemism of free-living terrestrial protists: phylogeographic assessment of the streptophyte alga *Klebsormidium*. *Environ Microbiol.*
- Škaloud P, Lukešová A, Malavasi V, Ryšánek D, Hrčková K, Rindi F (2014) Molecular evidence for the polyphyletic origin of low pH adaptation in the genus *Klebsormidium* (Klebsormidiophyceae, Streptophyta). *Plant Ecol Evol* 147 (3): 333-345.
- Škaloud P, Rindi F (2013) Ecological differentiation of cryptic species within an asexual protest morphospecies: a case study of filamentous green alga *Klebsormidium* (Streptophyta). *J Eukaryot Microbiol.* 60: 350-362.

### Diversity of diatom flora of polar streams

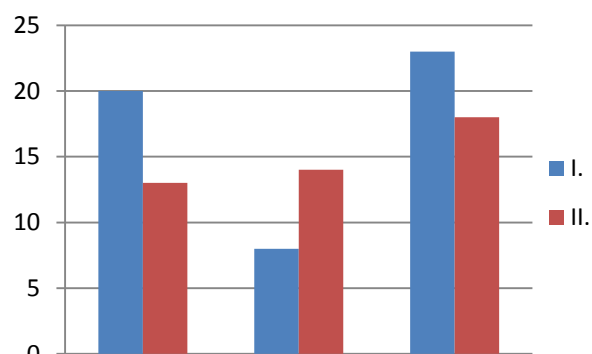
Terezie Englová compared diatom flora in different types of streams in Petuniabukta whence the first type were streams which were supplied by melting snow and the second type were streams which were supplied by glaciers.

There were selected two representatives of each type, glaciers streams were concretely supplied by Ferdinandbreen and the other by Ebbabreen. Diatoms were collected on three typical places: near their sources, next to marine terrace and near the sea in these selected streams. Collection of samples were done by a method of scraping periphyton from the whole surface of each stone and from three stone on each place. It were also measured some ecological variables like temperature of water, pH and conductivity. Then it was obtained empty frustules by a chemical method using nitric acid and determined species and their frequency based on Blanquet's scale.

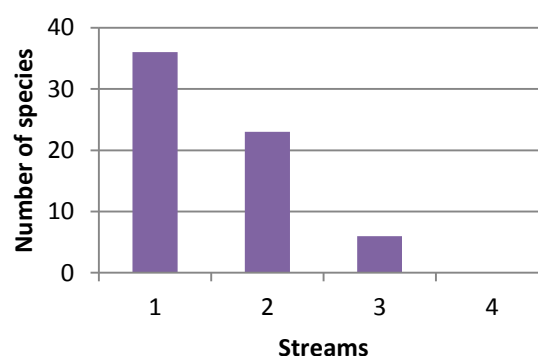
In streams which were supplied by melting snow the divergence was biggest near the sea which was probably caused by the slowest rate of flow (Fig. 3.1.8.).

It was found out, according to attached graph, that in glacier streams the diversity of diatom species were really low or none probably because of a big influence of abrasion of stones caused by entrained particles and big flow (Fig. 3.1.9.).

Additional experiments should be focused on a life of strategy of succesful species with high frequency (f.e.: *Hannaea arcus*, *Cymbella arctica*, *Gomphonema micropus*, *Gomphonema cf.*



**Fig. 3.1.8.** Diversity at each place in streams supplied by melting snow. Groups (from left): near source – intermediate part at terrace - near sea).



**Fig. 3.1.9.** Diversity of streams which were. 1 and 2 - snowmelt streams, 3 and 4 glacial streams.

*productum*...) but also on less frequent species which were typical for different spaces because these are the main bioindicators.