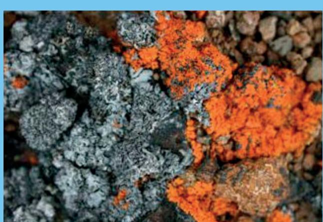
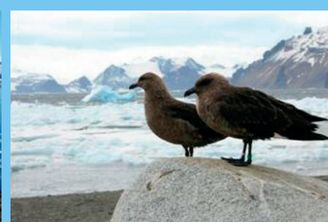
MASARYK UNIVERSITY

STRUCTURE AND FUNCTION OF ANTARCTIC TERRESTRIAL ECOSYSTEMS

Electronic Conference on Interactions between Antarctic Life and Environmental Factors, IPY-related Research
Brno, October 22th-23th, 2009



Book of Abstracts and Contributed Papers

Conference organised by
DEPARTMENT OF PLANT PHYSIOLOGY AND ANATOMY,
INSTITUTE OF EXPERIMENTAL BIOLOGY,
Faculty of Science, Masaryk University, Brno, Czech Republic

In co-operation with
UNIVERSITY OF SIENNA, ITALY
CZECH NATIONAL CENTER FOR POLAR REGIONS RESEARCH

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under the auspices of

Prof. PhDr. Petr Fiala, Ph.D., LL.M.
Rector of the Masaryk University
Assoc. Prof. RNDr. Milan Gelnar,CSc.
Dean of the Faculty of Science

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Contents:

Invitation:

STRUCTURE AND FUNCTION OF ANTARCTIC TERRESTRIAL ECOSYSTEMS

Electronic Conference on Interactions between Antarctic Life and Environmental Factors

International (Central European) electronic conference is focused on recent state-of-art in the research of structure and function of Antarctic terrestrial ecosystems. The scope of the conference includes: (1) STRUCTURE OF ECOSYSTEMS – abiotic factors, biotic factors, (2) HIGHER PLANTS - distribution, growth and production, interactions with physical factors, microclimate, ecophysiology, stress physiology, (3) LICHENS AND MOSSES – biodiversity and distribution, growth and production, interactions with physical factors, microclimate, ecophysiology, stress physiology, (4) TERRESTRIAL MICROBIOTA INCLUDING ALGAE AND CYANOBACTERIA – biodiversity, distribution, production, interactions with physical factors, ecophysiology, stress physiology, (5) TERRESTRIAL ANIMALS (VERTEBRATES) – population biology, etology, physiology, (6) TERRESTRIAL INVERTEBRATES – biodiversity, population biology, etology, physiology. The Conference will be held fully electronically at the Masaryk University web space. Contributions (4 pages each) will be peer-reviewed and published in The Book of Contributions and Abstracts. Abstracts will be published without any revision. The participant will be given a hardcopy of The Book of Contributions and Abstracts at the first day of the Conference. International prticipants will obtain the The Book of Contributions and Abstracts by surface mail.

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Program:

Thursday, October 22, 2009	Friday, October 23, 2009
10:00 Opening of the conference (Addresses of Masaryk University authorities)	9:00 Chat - open
10:00 Electronic Book of Abstracts and Contributions availible on the conference web page.	
11:00 Workshop of the delegates of the National Center of Polar Regions Research	21:00 Chat - closure

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Overview of Czech Ecological Research in Antarctica with a special Respect to the James Ross Island

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Though majority of the Antarctic continent is permanently covered with ice, there are number of ice- and snow-free places colonised by permanent vegetaion (mostly cyanobacteria, algae, moss and lichens). Due to their limited area and discontinuous character, such regions are sometimes denoted as *"Antarctic vegetation oases"*. Their development and permanent existence is determined by landscape morphology and local climate character that prevent permanent ice cover. The climate factors are prevailing in those particular areas where they cause delayed formation of snow layer and, consequently, ice (attenuated precipitation due to changed air advection conditions) to the extent that is exceeded by ablation rate and melted water flow.

Majority of Antarctic vegetation oases are found in the islands adjacent the west and north cost of the Antarctic Peninsula. This region is relatively easily accessible and a lot of research stations and facilities enabling long-term research of Antarctic vegetaion oases. This is particularly true for the *King George* island where some of the largest and richest are situated, especially in the vicinity of the Polish research base *Henryk Arctowski.* Several Czech experts, particularly algologists and climatologists were involved in the research there in 1980s and 90s. More intensive Czech involvement in AVO research has been possible since 1998 thanks to executing the research project named *"Ecology of Antarctic Vegetation Oasis"* (RP J07/98:143100007) by the Masaryk University. Fieldwork has been performed in other AVOs in the vicinity of the Peruvian research station (*Crepin Point*, *King George Island*) and at the Ukrainian station Vernadsky (*Galindez* island, *Argentine Islands*) since then.

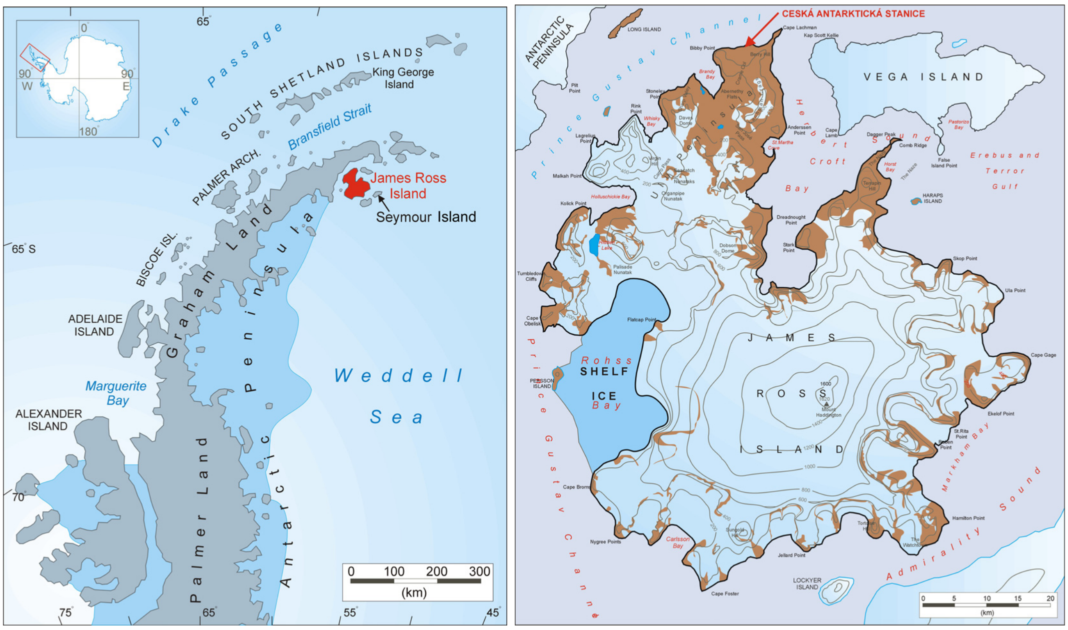

Fig. 1. Location of the James Ross Island and J.G.Mendel station

The idea to build a Czech seasonal research station in the Antarctica originated from the experience gathered through activities of Czech scientists in the area of the Antarctic Peninsula and South Shetlands, and from the need to establish a logistic background for development of complex and long-term research. The first proposed site of the station was located at the Turret point, east part of King George Island (South Shetlands). After comments on the first proposal of the station site presented at the XXIVth ATCM in St Petersburg, 2001, a new location was proposed at the northern coast of the James Ross Island.

In 2006, Czech Antarctic station J.G. Mendel was established at the northern seashore of the James Ross Island (φ = 63°48′02′′S; λ = 57°52′57′′W, see maps). Building of the station construction was supported by the funding provided by the Czech Ministry of Education, Youth and Sports within the project (INGO LA 118) "Building a Czech station in the Antarctic". The project was carried out by the Masaryk University, Brno within the period of January 2000 – March 2006. Technical project and construction of the station was developed by Investprojekt, ltd. Zlín, in collaboration with Ecosolaris Kroměříž, the Czech Technical University Prague, and the Technical University Brno. The chief construction contractor was PSG International Zlín. Subcontractors were: CZECH PAN ltd. Varnsdorf (wooden structures); Elma-therm ltd. Kroměříž (electrosystems); AZ KLIMA ltd. Brno (air-conditioning); MG PLAST ltd. Letovice (wind energy); VLW ltd. Zlín (diesel-powered generators); Ct ltd. Komárno, Slovakia (containers); Marine Equipment ltd. Nové Město nad Metují (ship equipment); Ekosolaris Kroměříž (solar equipment); and CSM Tisovec (off-road vehicle). The shipping was organised by Czechoslovak Ocean Shipping ltd.

Czech station now consists of main operation building and 9 independent technical containers, each of which is equipped with wind power station. Main operational building is a wooden construction with efficient thermal insulation. The station is heated by solar air heating with supplemental heating provided by electricity produced mainly by eight wind-powered electric generators with a total output of 12–12 kW. Backup electrical supply is secured by a diesel generator. The technical needs of the station are supplemented by nine independent containers which comprise electrical energy unit, storage of spare parts, fuel, food, the incineration of waste, and a garage for vehicles. The J.G. Mendel station provides safe accommodation for 15 people.

Since 2006, majority of the Czech Antarctic research has been located to the James Ross Island ad surrounding areas of the East coast of the Antarctic peninsula. Recent main research directions might be summarized as follows:

(1) Continuous monitoring and evaluation the *deglaciation process* of the costal zone in the *James Ross* Island with a special respect to physical, geologial, and geographical aspects. Another goal is monitoring of periglacial processes focused at rock material transport, deglaciated region modelling and permafrost surface layer changes.

(2) Continuous monitoring and analyses the *spatio-temporal processes of climatic character* (including radiation and energy balance) leading to formation of specific local climate, microclimate and soil climate in the vegetation oases. Monitoring of UV radiation and its components observing their regime while considering atmospheric factor effects on the western and eastern side of this orographic barrier.

(3) Quantification the *pedochemical and hydrochemical processes,* seasonal and long-term dynamics. Special attention is paid to mineral nutrient release by rock

weathering and to formation of soil organic mass including humic substances synthesis processes. Detailed investigation of lakes located in the northern part of the James Ross Island.

(4) Quantification of the *processes related to the nitrogenous compounds circulation* in the vegetation oases ecosystems including their transformation and efficient utilisation in the ecosystems biological components and definition of significance for each of the transport routes.

(5) Monitoring of the *biotic colonisation process* in the new deglaciated regions and observation of changes in biodiversity and plant community composition in a series of sites exposed to different environment factors.

(6) Estimation of the carbon fixation, storage and release related to *physiological processes* (photosynthesis, respiration) rates in significant components of vegetation, and analysis of the processes as dependent on external environment factors.

(7) Investigation of mechanisms involved into *stress physiology* of typical representatives of autotrophic Antarctic organisms. Study of background of adaptations to extreme environments.

(8) Long-term controlled field experiments aimed to manipulated environment approach. Main manipulation is temperature increase caused by open top chambers installed in typical ecosystems of the james Ross Island. Long-term experiment is aimed to predict the likely changes in biomass production, community structure and biodiversity of Antarctic vegetaion oases under global climate change.

(9) Collection of basic cartographic and/or geoinformatic data so that detailed geological and geomorphological map of the James Ross Island can be constructed.

Recent research projects that were included into the 2008/2009 expedition to the James Ross Island cover several fields: glaciology, geology, geomorphology, paleochemistry, geography, climatology, limnology, ecology, ornithology, botany, vegetation analyses, long-term experiments with manipulated air temperature and its effect on lichens and mosses, plant stress physiology. Recently running projects are:

(1) Project VaV660/1/03: Program of Antarctic Research (Czech Geological Survey)

(2) Recent deglaciation of the northern part of James Ross Island, Antarctica (GA205/09/1876, Masaryk University, Czech Geological Survey)

(3) The effect of atmospheric factors on the regime of UV radiation in the region of Antarctic Peninsula (GA205/07/1377, Masaryk University)

(4) Multidisciplinary ecological research of Antractic terestrial vegetation within the IPY (ME 945 - KONTAKT), Masaryk University, South Bohemian University

Fig. 2. Initial phase of construction of J.G.Mendel station building (2006).

Fig. 3. J.G.Mendel station in 2007/2008 austral summer season

Meteorological measurement and observation at the J. G. Mendel Station, James Ross Island, Antarctica

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Introduction:

Recently, rapid climate warming along the Antarctic Peninsula has been reported by many authors. It is generally known that changes of atmospheric circulation have the largest impact on positive trend of near surface air temperature in this region. Geographical location of the J. G. Mendel Station on the northern ice-free part of the James Ross Island (situated only 15 km off the east coast of the Antarctic Peninsula) allows to study the impacts of regional climate change on the terrestrial ecosystems.

The main goal of meteorological measurements and observations at the J. G. Mendel Station and its vicinity is to 1) investigate the effects of atmospheric factors on the spatiotemporal variation of the UV radiation, 2) evaluate recent deglaciation processes, their responses and sensitivity to rapid regional climate warming, 3) investigate the response of lichen and moss vegetation to manipulated warming environment using long-term installations of the open top chambers, and 4) study the constituents of terrestrial ecosystems, especially their responses to low and freezing temperature, limitations in water availability and solar radiation stress.

Material and Methods:

At present, monitoring network consists of nine automatic weather stations (here after AWS) situated at different altitudes ranging from the seashore level up to mesas and glaciers, respectively, located hundreds meters above sea level. All sites are in open environments with little or no influence of local topographical factors and terrain inter-shielding. The first AWS was installed on the northern coast of the James Ross Island (Ulu Peninsula) in February 2004. It was located on marine terrace situated about 150 m southward from the main building of the J. G. Mendel Station (see Fig. 1). Extended measuring program on this AWS consists of the measurement of incident and reflected global solar radiation, air temperature and relative humidity at heights of 2 and 11 m, wind speed and direction at 10 m, atmospheric pressure, surface temperature, soil heat flux, and soil temperatures at depths of 5 to 150 cm. A special attention is devoted to monitoring of solar radiation and its characteristic components, in particular UVA, UVB and global UV radiation, erythemally effective UVB, and photosynthetically active radiation. There are the following instruments installed at a special platform on the top of a technical container: CM11 pyranometer, CUV3 and UVSABT radiometers manufactured by Kipp and Zonen (Netherlands), UV-Biometer Robertson Berger Model 501A (Solar Light Co., USA), PAR sensor (Environmental Measuring Systems, Czech Republic). Apart from that, additional six AWSs were established at different locations with specific environmental conditions in the period of 2005-2007. At all localities, there are measured basic meteorological parameters such as global solar radiation, air temperature and relative humidity, near surface ground temperatures at depths of 5 to 30 cm.

Results:

The climate conditions of the northern part of the James Ross Island are affected by the regional-scale atmospheric circulation formed by the Antarctic Peninsula mountains and advection of different (maritime and cold continental) air masses. Prevailing winds at the J. G. Mendel Station are from the sector ESE-SSW, while the NNW-N winds occur only rarely.

Fig. 1. View on the meteorological tower with instruments at the J. G. Mendel Station.

The climate of the Ulu Peninsula is typical of a short summer (December-February) with air temperature fluctuation between -5 and 10°C. The annual mean temperature at the J. G. Mendel station is -5.4 °C. The warmest month is January, while the coldest one is July and August. Winter air temperatures show large interdiurnal variability because of cyclonic activity along latitudes 50°S and 60°S and sudden advections of different air masses. The mean annual sum of global solar radiation reaches up to 3800 $MJ.m^{-2}$. The minimum monthly sum of radiation occurs in July. However, maximum daily sum of global solar radiation can be reached as high as 35 $MJ.m^{-2}$ on clear sky days around summer solstice. Large diurnal and interdiurnal variability of solar radiation fluxes at the J. G. Mendel Station is affected mainly by cloudiness and cloud genera linked up with cyclogenesis over the Antarctic Peninsula.

Acknowledgments: This research is funded by the project of the Czech Grant Agency 205/09/1876.

Glacio-meteorological measurements on two glaciers at the northern part of James Ross Island, Antarctica

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Introduction:

Antarctic Peninsula has been one of the most rapidly warming parts of our planet during the last haft of the century. The consequences of this climate change are clearly documented by recent disintegration of ice shelves on both sides of the Antarctic Peninsula as well as by the retreat of land-based glaciers. Therefore, the James Ross Island located close to the northernmost tip of the Antarctic Peninsula, represent an excellent place to study changes in the glacier mass-balance and their sensitivity to regional warming trend. This is particularly true for its northern part – the Ulu Peninsula.

Basic description:

We selected two different types of glaciers for this study. ***Whisky Glacier*** is a well-delimited valley glacier recently lying mostly below the local Equilibrium line altitude (ELA) with high flow velocities. The Whiskey Glacier is fed by an intensive snow accumulation caused by prevailing southwestern winds. The Whisky Glacier has an area of 2.306 km^2 and lies in the altitude range of 215–475 m a.s.l. (summer 2006 situation). ***Davies Dome*** is a higher lying flat-bottom dome glacier with significant part of its surface located above the ELA and only limited flow velocities in most of its body. However, the Davies Dome has a single 500–700 m wide southwestern outlet flowing towards the Whisky Bay. The Davies Dome has an area of 6.693 km^2 and lies in the altitude range of 0–514 m a.s.l. (summer 2006 situation). Both glaciers experienced massive extension of their ice tongues towards the Brandy Bay during the mid Holocene. This is expressed by lateral moraine ridges located in front of both glaciers heading down to the left coast of the Brandy Bay (see Fig. 1).

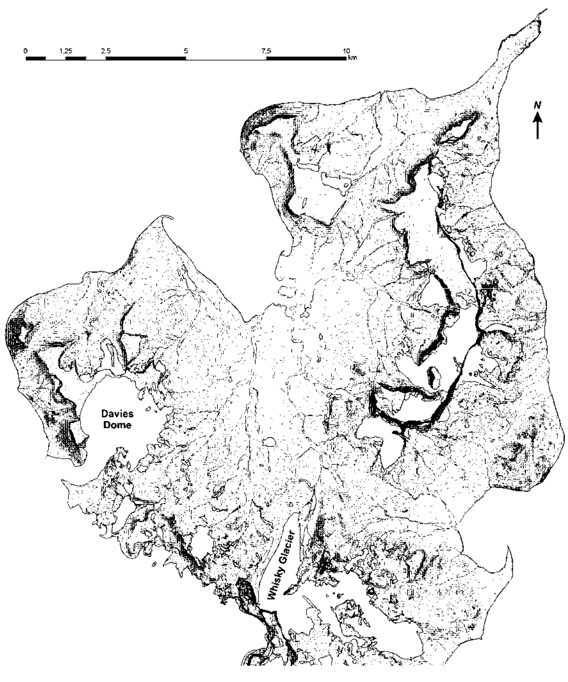

Fig. 1. Location of the Davies Dome and Whiskey Glacier in the northern part of the Ulu Peninsula, James Ross Island.

Results and Discussion:

The first meteorological and glaciological observations started on Davies Dome in January 2006. In the austral season of 2009, two automatic weather stations (AWS) were installed on both glaciers. Each AWS was equipped with albedometer CM7B (Kipp-Zonen, Netherlands), air temperature and humidity sensor EMS33 (EMS, Czech Republic), propeller anemometer 05103 (Young, USA), and snow depth sensors (Judd, USA). Furthermore, annual mass balance method based on the measurements of ablation stake heights, snow density and stratigraphy were applied on the glaciers. We established first transect of 24 ablation stakes with 4 control points on Davies Dome in January 2006. This was complemented in the austral season of 2009 by other 19 ablation stakes on Davies Dome and a new network of 20 stakes installed on Whisky Glacier (see Fig. 2). The annual mean temperature on the top of Davies Dome was -9.3 °C in the period of 2007–2008. The sum of positive degree-day temperature (number of days with daily mean temperature >0 °C) varied from 79.2 °C d in 2007 to 108.3 °C d in 2008. Mapping of 2-D extent of Davies Dome between 2006 and 2009 revealed glacier margin retreats especially in the NW flat part in order of 10–20 m. There are also clear changes in ablation along the transect over the Davies Dome, with the average ablation in order of 20 cm between the years 2006 and 2009.

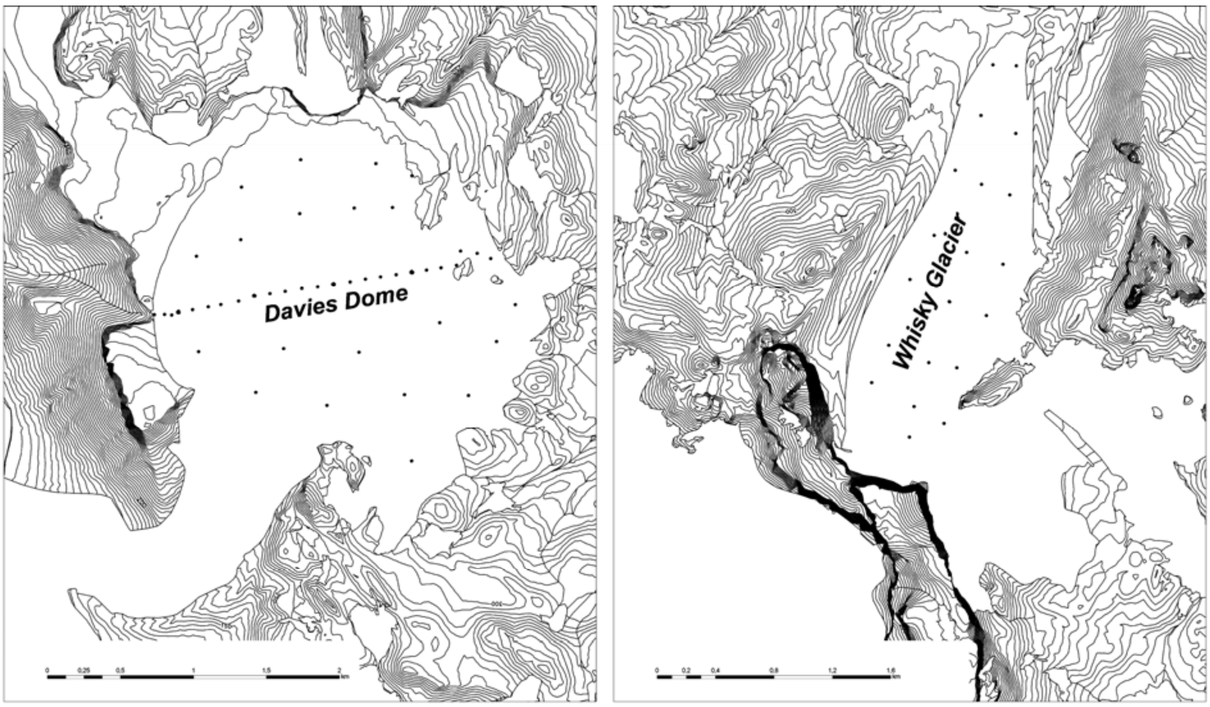

Fig. 2. Location of the ablation stakes networks on Davies Dome and Whisky Glacier.

Acknowledgments: This research is supported by the project of the Czech Grant Agency 205/09/1876 and by the R & D project VaV SP II 1a9/23/07.

Pedobiology studies in Hasswell Archipelago, East Antarctica

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Introduction:
This work aims to determine the soil forming level reached by the terrestrial crust in this area of extreme climate and its modification under climate changes. In the Antarctic Continent, because of the harsh climate conditions, the soils are generated as shallow layers, in small depressions at the shelter of the rocks, from the mineral material resulted from physical processes (terrestrial crust weathering), where under favourable climatic conditions the microflora generated the first organic material. (Beyer *et al.* 1999, Negoita T. Gh., Stefanic Gh. et. al. 2001, 2002). The differentiation of the incipient soils from the defined ones may only be done by determining the occurrence of vital and enzymic processes, as well as the biomass and humic substance content accumulation. It is very likely that the microfauna also contribute to the biomass increase in the incipient soils. Results in this study belong to the IPY Project 1267 "Pedobiological processes evolution in polar areas" (PPEPA), proposed by Negoita T. Gh.

Material and Methods:
Soil samples were separately and aseptically gathered in 2006 by Negoita from depths of 0-9 cm and 9-12 cm. Microbiological analyses involved determining the saprophyte bacteria number (colony forming units cfu) by soil decimal dilution method, and the soil granule colonization index with micromycetes as per the Parkinson *et al*. method (1969), on the water-agar-valerian extract medium (Papacostea 1976). Soil respiration potential was determined by Stefanic respirometer (1991, 1994). The cellulosolythic potential was tested by Vostrov&Petrova method (1961), improved by Stefanic (1994). Enzymic analyses: Soil catalase potential was tested by an automate catalase-meter (Stefanic *et al.*1984), by determining the released oxygen (cm^3/100g soil d.s./min) from a solution of 10% H_2O_2. Saccharase potential was determined by a spectrophotometric method (Stefanic 1972) estimating the hydrolysed saccharose quantity; the total phosphatase potential by Irimescu&Ştefanic (1999) method. Chemical analyses were performed by classical methods.

Results and Discussion:
Our research put into evidence the very high contents of: organic carbon in soils from Mirny zone (Hasswell Archipelago), i.e. 2.33 % on an average, total nitrogen, 0.595 % and organic phosphorus, 0.42 mg P/100 g soil d.s. (as compared with 0.07-0.16 mg P in the other stations), also averages respectively, fact that points out good biological conditions for the mineralization processes and the organic matter instability in relation with the mineralizing activity of microflora, due to its prevailing content consisting of penguin and skua dejections. The differentiation between mineral and organic soil was done by determining the occurrence of vital and enzymic processes, as well as biomass and humic substance content accumulation. The analytical values concerning the potentials of respiration and cellulolytic activities-Indicator of Vital Activity Potential % and the potentials of catalase, saccharase and total phosphatase - Indicator of Enzymic Activity Potential % and the Biologic Synthetic Indicator (BSI % = (IVAP % + IPAE %) : 2, were taken into consideration. Corresponding to the methodology (Ştefanic 1994, 1999 and 2001) the determination of biological potentials of soil samples is to be achieved in laboratory standard conditions and, generally, the more the values of indicators increase, the more one considers that the soil is more developed.

Acknowledgements: Supported by Romanian Antarctic Foundation, 2007.

Diversity of microfungi from James Ross Island, Antarctica

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Key-words: Antarctica, filamentous fungi, black fungi, psychrophilic organisms

Introduction:

The Antarctic mycobiota is diversified depending on differences in local climate and substrate. Presence of bird colonies, invertebrate populations and vegetation as well as geographic location affects the occurrence and frequency of microscopic fungi. Most Antarctic filamentous fungi represent common mesophilic species widespread in the biosphere, capable of adaptation to stress conditions such as low temperatures, high UV radiation, low water and nutrients availability and repeated freeze and thawing cycles [7]. However, few Antarctic fungi are true psychrophilic organisms such as some species of *Thelebolus* [2], cryptoendolithic black meristematic fungi [9], *Chaunopycnis ovalispora* or *Acremonium psychrophilum* [4, 5]. Some of them are most probably endemic (e.g. *Friedmanniomyces* [6] and *Cryomyces* [9] or *Thelebolus ellipsoideus* and *Thelebolus globosus* [2]). On the other hand some species are frequently recorded from various sites and substrates in Antarctica including *Geomyces pannorum*, *Phoma herbarum* and *Thelebolus microsporus* [7].

In this study, we focused on microfungi occuring in ice-free terrestrial ecosystems of the Ulu Peninsula (Northern part of the James Ross Island). Soil crusts represent one of important components of the area of interest and along with phyto- and zooedaphon in the active layer form a complex mosaic of microhabitats. The purpose of this project is to study the diversity of filamentous fungi from soil crusts and other substrates of different Antarctic ecological niches.

Material and Methods:

The investigated area is located at the ice-free part of the James Ross Island, nearby the Czech Antarctic station of Johann Gregor Mendel (60°48´2.3″S; 57°52´56.7″W). About 80 % of the island surface is covered with glacier. The entire ice-free part of the Ulu Peninsula (150 km^2) is covered with consistent permafrost. During austral summer, the active layer of permafrost is located in the depth of 40-80 cm in the majority of ice-free area of the Ulu Peninsula. The vegetation of the island is represented by cryptogamic tundra, with mosses and lichens as the main components of communities forming Antarctic vegetation oases.

A total of 67 samples of soil crusts and other substrates were collected in 12 habitats with different microclimate and water availability during three austral summers of 2007 - 2009. Among them, the following were most frequent: (1) north-faced sunny slopes of the Berry Hill (see Fig. 1), (2) shady site near the snowfield located at the south-eastern foot of the Berry Hill (Fig. 2), (3) permanent wetting sites and (4) places inside bird colonies. Soil samples were collected aseptically into sterile polyethylene bags or tubes and stored at -20 °C. During transportation from Antarctica to the Czech Republic, a portable fridge and cooling bags assuring temperature below 5 °C were used.

Filamentous fungi were isolated by dilution method and direct plating of sample fragments on Petri dishes containing MEA (Malt Extract Agar, HiMedia) with chloramphenicol and DRBC (Dichloran Rose-Bengal Chloramphenicol Agar, Merck). Plates were incubated in dark at 10 and 15 °C. Development of new fungal colonies was checked weekly during one month. Individual colonies were subcultured on MEA in order to obtain pure cultures. Identification to species or genus level was based on microscopic and macroscopic characters using diagnostic media [1, 3, 8] and slide culture methods [1].

Fig. 1. Sampling site - sunny slopes of the Berry Hill.

Fig. 2. Sampling site - shady site near the snowfield located at the southeastern foot of the Berry Hill.

Results and Discussion:

Totally, 286 isolates of filamentous fungi were obtained from 67 samples, 161 of which were identified as 25 species within 17 genera. The remaining 125 isolates representing Hyphomycetes, Coelomycetes, Agonomycetes, Basidiomycota and subphyla incertae sedis formerly classified as Zygomycota have not been identified yet. List of all identified species and their frequency of occurrence in relation to the total number of analyzed samples is shown in Table 1.

The most frequent taxa were *Geomyces pannorum* var. *pannorum*, *Phoma herbarum*, *Mortierella* spp., *Cladosporium* spp. and *Penicillium* spp. *Penicillium brevicompactum* and *Cladosporium herbarum* were the most common species of *Penicillium* and *Cladosporium*. *G. pannorum* var. *pannorum* was isolated from all sampling sites except one. This species is frequently isolated in Antarctica from various sites and substrata [7]. *P. herbarum* was present in 9 of 12 sampling sites. This fungus is recorded as the most abundant species on stems of mosses [10]. However, occurrence of some fungi was limited. Species with percentage occurrence 1,5 were recorded from a single sample. Some fungi live in association with other organisms. *Beauveria bassiana* isolated from 4 sampling sites and *Isaria farinosa* found in one sample are known to be entomoparasitic. Most of these fungi have been reported in previous mycological studies in Antarctica [2, 5, 10, 11]. In contrast *Engyodontium rectidentatum, Penicillium miczynskii, Spiniger meineckellus* and *Verticillium leptobactrum* were isolated from samples collected in Antarctica for the first time in this study.

The majority of the filamentous fungi listed in Table 1 are cosmopolitan, cold-tolerant mesophilic species. They are capable to grow and reproduce at least under Antarctic summer conditions. Moreover, a few of them are psychrophilic including *Thelebolus microsporus* and black meristematic fungi. Black melanised fungi exhibit meristematic growth and form compact restricted microcolonies inside rocks [9]. Group of seven strains of black meristematic fungi were isolated only from cold habitats of the Ulu Peninsula. Molecular studies in cooperation with CCFEE (Culture Collection of Fungi from Extreme Environments) to assess their phylogenetic position are still in progress; preliminary data address all these fungi to the order *Capnodiales*. *T. microsporus* is frequently reported from Antarctica in association with Antarctic bird dung and feathers [2]. In our study, the high number of *T. microsporus* isolates was obtained from soil inside bird colonies. Most of the isolated fungi represented anamorphic forms. Only *T. microsporus* reproduced sexually. The results of mycological analysis revealed relatively high species diversity in soil crust in spite of stressful conditions influencing their development. The mycobiota of James Ross Island is mainly formed by anamorphic Ascomycota, only a few species of Basidiomycota and Mucoromycotina were present.

Table 1. List of identified species and their frequency of occurrence in relation to the total number of analyzed samples.

Species	%	Species	%
Acremonium bacillisporum	1,5	*Mortierella antarctica*	1,5
Acremonium butyri	3	*Mortierella* spp.	19,4
Acremonium sp.	1,5	*Onychophora* sp.	3
Acrodontium cf. *crateriforme*	4,5	*Penicillium aurantiogriseum*	1,5
Arthrinium sp.	1,5	*Penicillium brevicompactum*	6
Aspergillus versicolor	1,5	*Penicillium chrysogenum*	1,5
Beauveria bassiana	6	*Penicillium spinulosum*	1,5
Cladosporium cladosporioides	3	*Penicillium miczynskii*	3
Cladosporium herbarum	7,5	*Penicillium roqueforti*	1,5
Cladosporium sphaerospermum	1,5	*Penicillium* sp.	1,5
Cladosporium spp.	10,5	*Phoma herbarum*	25,4
Engyodontium album	1,5	*Rhizoctonia solani*	1,5
Engyodontium rectidentatum	3	*Spiniger meineckellus*	1,5
Fusarium cf. *sporotrichioides*	1,5	*Thelebolus microsporus*	10,5
Geomyces pannorum var. *pannorum*	38,8	*Verticillium leptobactrum*	6
Isaria farinosa	1,5	Black meristematic fungi	7,5

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Invited paper

Two freshwater filamentous, branched green algae, newly discovered in Antarctica.

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The members of freshwater and terrestrial filamentous green algae with branched thallus (the traditional order Chaetophorales), which is diversified according to modern taxonomical criteria in various clades and classified in different taxonomic families, have been recorded mostly from the subantarctic islands and regions, in which are developed also communities of vascular plants (e.g., Southern Patagonia, Kerguelen Islands, Orkney Islands, Falklands – *cf.* Prescott 1979). From such habitats few species are recorded (Tab. 1). Contrastingly, no confirmed locality of such algae exists from maritime and continental Antarctica.

Our phycological activities in 2009 on the northern part of James Ross Island (NW Weddell Sea) were oriented to the investigations of the diversity of green algae and cyanobacteria. In lakes of NE part of this island, two species were found from the traditional order Chaetophorales, which represent evidently new, not yet known types. The location of lakes is presented in Fig. 1. The main ecological and morphological characteristics of lakes with both new algae are as follows (*cf.* Tab. 2):

1. The Lachman lakes represent a system of two connected shallow water bodies, between of which a system of small puddles and pools exists (Fig. 2). In the periods of higher water level (after melting of snow at the beginning of Antarctic summer season), the both lakes can have their own outflows to the sea, but their connection is also possible. The lakes are localized on the sea terrace, approximally 30 m above sea level. During the summer season, the water level slightly decreases.

   During the summer season (from November), massive cyanobacterial mats develop locally in shallow littoral parts of the lakes . From the beginning of January, the occurrence of a special type of *Stigeoclonium* sp. starts. The species grows attached to the littoral stones on east shore of the lake Lachmann II and in several pools located in the seepages between the both lakes, from which the water flows into the lake. The algal vegetation of typical *Stigeoclonium* filaments with sporadical branches begins in the middle of summer season (January), and changes during few weeks (to the middle of February) in massive clusters of richly branched colonies (Fig. 3). The vegetation terminates at the end of February and ends with the decrease of temperature below 0^{o}C with colonies disintegration.

   After collection in the field, *Stigeoclonium* colonies were transferred in monospecific cultures. Their morphology, life cycles and position in phylogenetic tree were studied (Fig. 4). From the preliminary results follows, that the Antarctic populations belong to the cluster containing the type species of the traditional genus *Stigeoclonium* (*S. tenue*) and that it does not correspond to any known and up to date described species (cf. Islam 1963, Starmach 1972, etc.). It must be therefore described as a new species and as the preliminary name is proposed "*Stigeoclonium bartakii*" nomen provis. (named to the honor of the chief of the Czech Antarctic expedition in James Ross Island in the year 2009, who first discovered and collected material of *Stigeoclonium*).

2. Green Lakes are situated among hills near Andreassen Point, approximately 8 km south from Lachmann Lakes, and their characteristics are slightly different (Fig. 5). The water supply changes during the summer season. In the flat littoral, which remains sometimes above water level, a very special community of autotrophic microorganisms with dominant

cyanobacteria *Calothrix* sp. and subdominant *Coleodesmium* sp. develop on stones. Such community participates in formation of special small, ridge-like stromatolites (Elster et al. in press). On the top of such stromatolites, a green chaetophoralean alga develops during Antarctic summer season, forming very dense, pillow-like, several mm high colonies (Fig. 6). The morphology is characterized by a special type of branching, life cycle, variability in cell form and by formation of a special asexual reproduction cells.

The population was transferred in a monospecific culture. Several morphological characters, life cycle and the phylogenetic status (Fig. 6) were verified from the culture. The classification in chaetophoralean algae was proved, but the status of a special new genus within this group was indicated.

Preliminary, the ecologically and morphologically new type is very interesting. The new type was designated as *Andreassenia antarctica* gen. et spec. nova, nomen provis. The name of alga was proposed according to the locality of collection: both the lakes of its occurrence are located in the region, which belong to the Andreassen Point, named after steersman of the famous Nordenskiöld expedition to James Ross Island.

Summary:

The discovery of two new species of chaetophoralean algae in Antarctic lakes is important for the knowledge of diversity of this group of algae over the world, and also for the understanding of development of Antarctic microflora. The interesting fact is that both localities (lakes) belong to geologically oldest parts of the James Ross Island, which are located in the NE part of the island. The age of Lachman Lakes is about 9000 years in comparison with lakes in other parts of the island, the age of which is estimated to about 2000 years (e.g., the Monolith Lake, inland of the north part of James Ross Island) (Ingólfsson et al. 1992, Björck et al. 1996). The Green Lakes near Andreassen Point are situated also in the geographically old region, similarly to Lachman Lakes.

Both species of more or less taxonomically problematic freshwater green algae prove the specificity of Antarctic microflora, which was sometimes considered as mainly secondary. Particularly the special morphology and ecology of *Andreassenia antarctica* indicate the special adaptation and development of this type under extreme Antarctic conditions. The detailed description of both types and the taxonomic validation will be published in next paper.

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Table 1. Filamentous branched green algae (Chaetophorales) recorded from Antarctic region. Citations according to Prescott (1979).

Chaetophora elegans	S Patagonia (Borge 1901)
Draparnaldia distans	Kerguelen Isl. (Reinsch 1879, Hemsley 1885, Wille 1924, Hirano 1965)
Draparnaldia glomerata	Kerguelen Isl. (Gain 1912, Therezien 1976); S Patagonia (Borge 1901)
Draparnaldia pusilla	Falklands (Hooker 1947)
Draparnaldia subtilis	Kerguelen Isl. (Reinsch 1876, 1879, Hemsley 1885, Gain 1912)
Gomontia arhiza	Tierra del Fuego (Hariot 1892)
Stigeoclonium falkandicum	Falklands (Gain 1912, Hirano 1965); Kerguelen Isl. (Bourrely & Manguin 1954, Hirano 1965, Therezien 1976)
Stigeoclonium hookeri	Kerguelen Isl. (Reinsch 1876, 1879, Hemsley 1885)
Stigeoclonium subtile	Kerguelen Isl. (Reinsch 1876, 1879, Hemsley 1885)
Stigeoclonium tenue	S Patagonia (Borge 1901)

Table 2. Summary of ecological data (summer averages) of the lake Lachman II and the Green Lake from mountainous part near Andreassen Point (acc. to Elster et al., in press).

Lake	depth [m]	temperature [^{o}C]	pH	alkalinity [mmol.L^{-1}]	conductivity [µS.m^{-1}]
Lachman II	0.3	11.0	7.3	288	118
Green L.	0.3	9.4	7.2	236	55.4

Fig. 1. Map of north part of James Ross Island with studied localities in Lachman Lakes and Green Lakes.

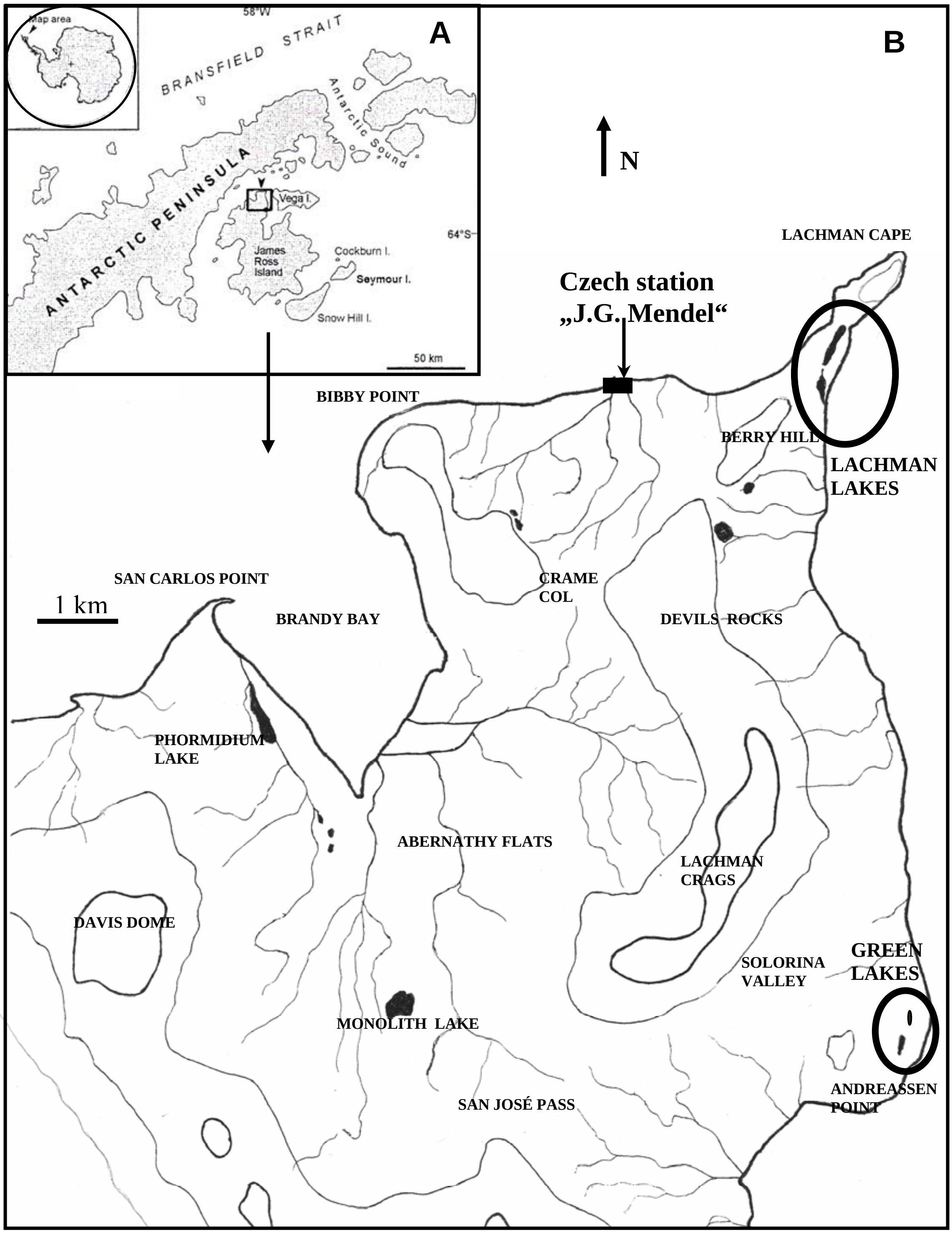


Fig. 2. Lachman Lakes with localities (white arrows) of *Stigeoclonium bartakii* (nomen provis).

Fig. 3. Colonies of *Stigeoclonium bartakii* in natural habitats.

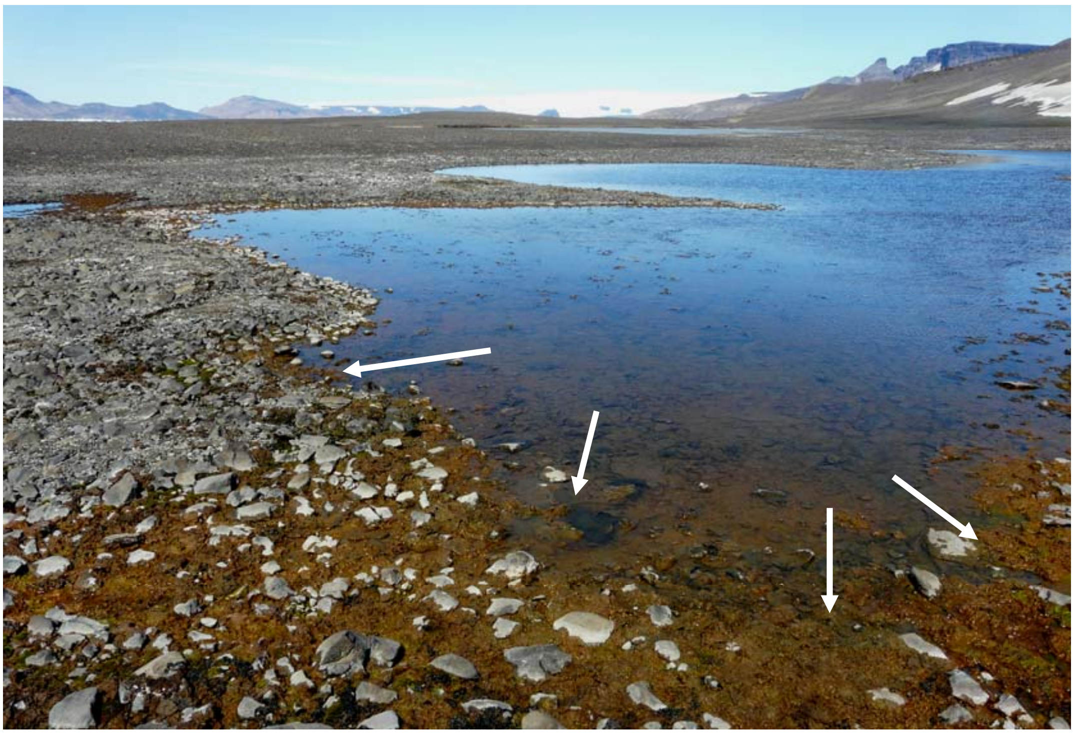


Fig. 4. Phylogenetic tree of Chaetophoralean green algae with the preliminary position of *Stigeoclonium bartakii.*

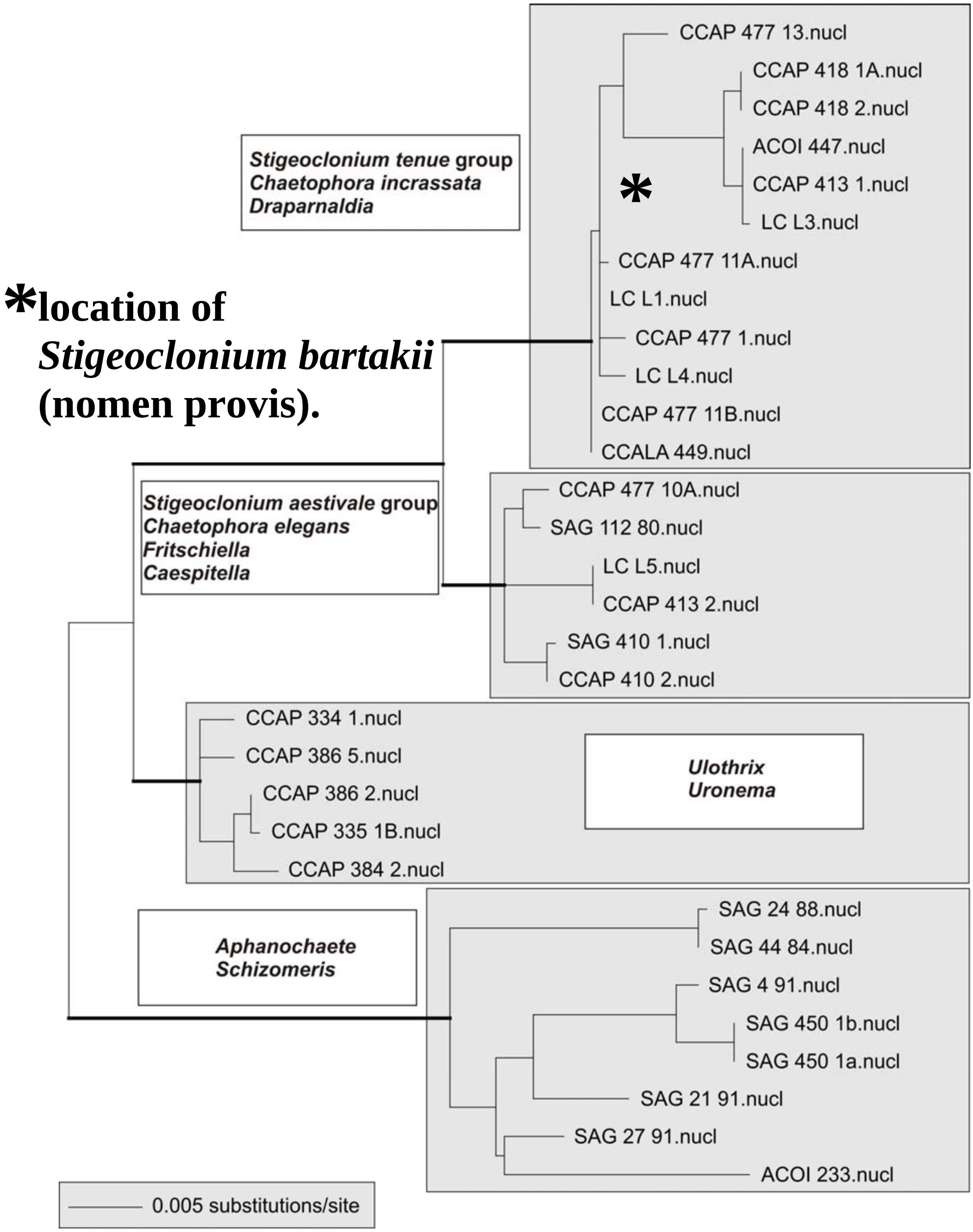


Fig. 5. Green Lake (A) and New Green Lake (B) near the Andreassen Point.

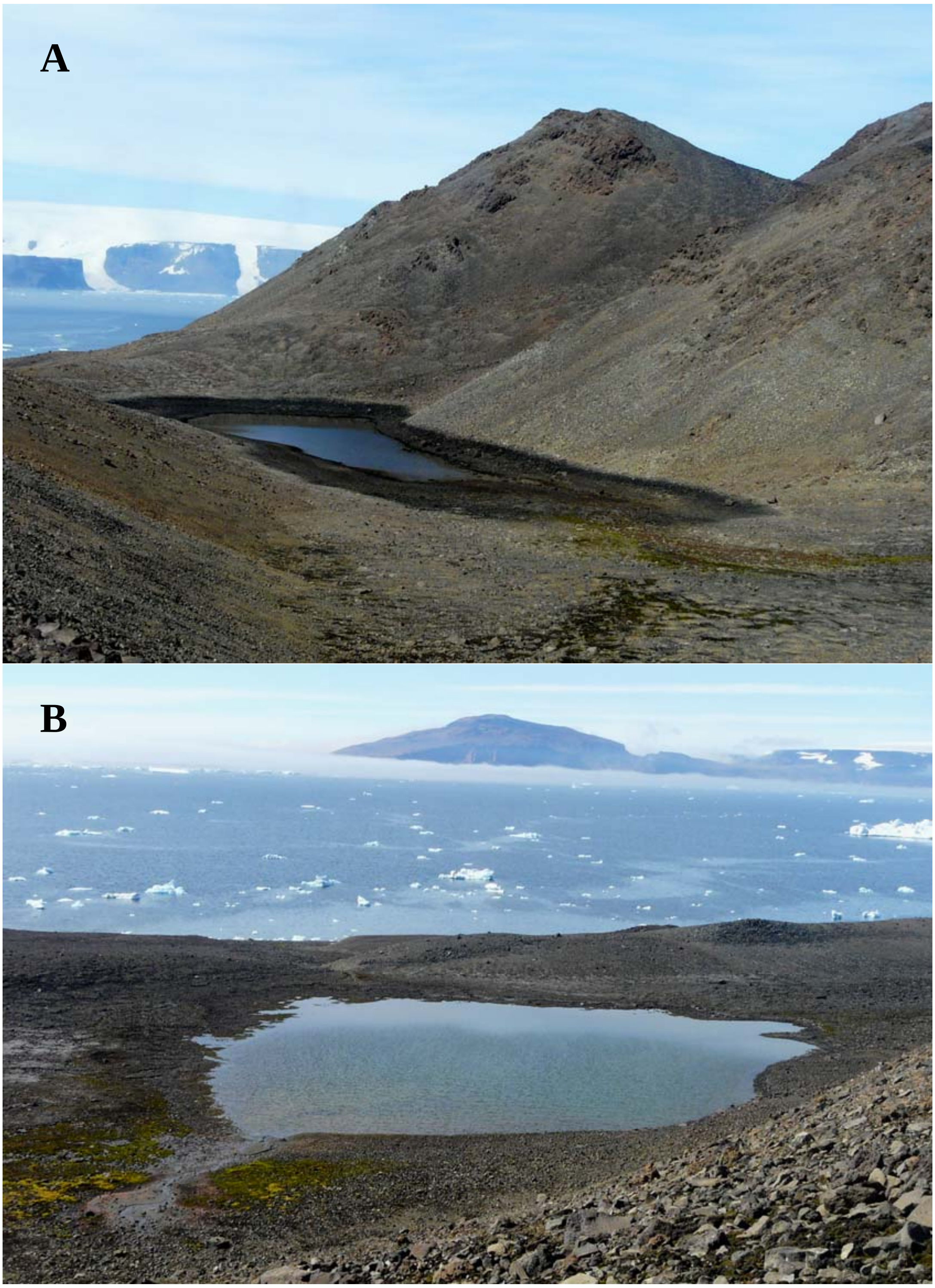


Fig. 6. Surface of stromatolites from Green Lake (A) and New Green Lake (B) with developed populations of *Andreassenia antarctica.*

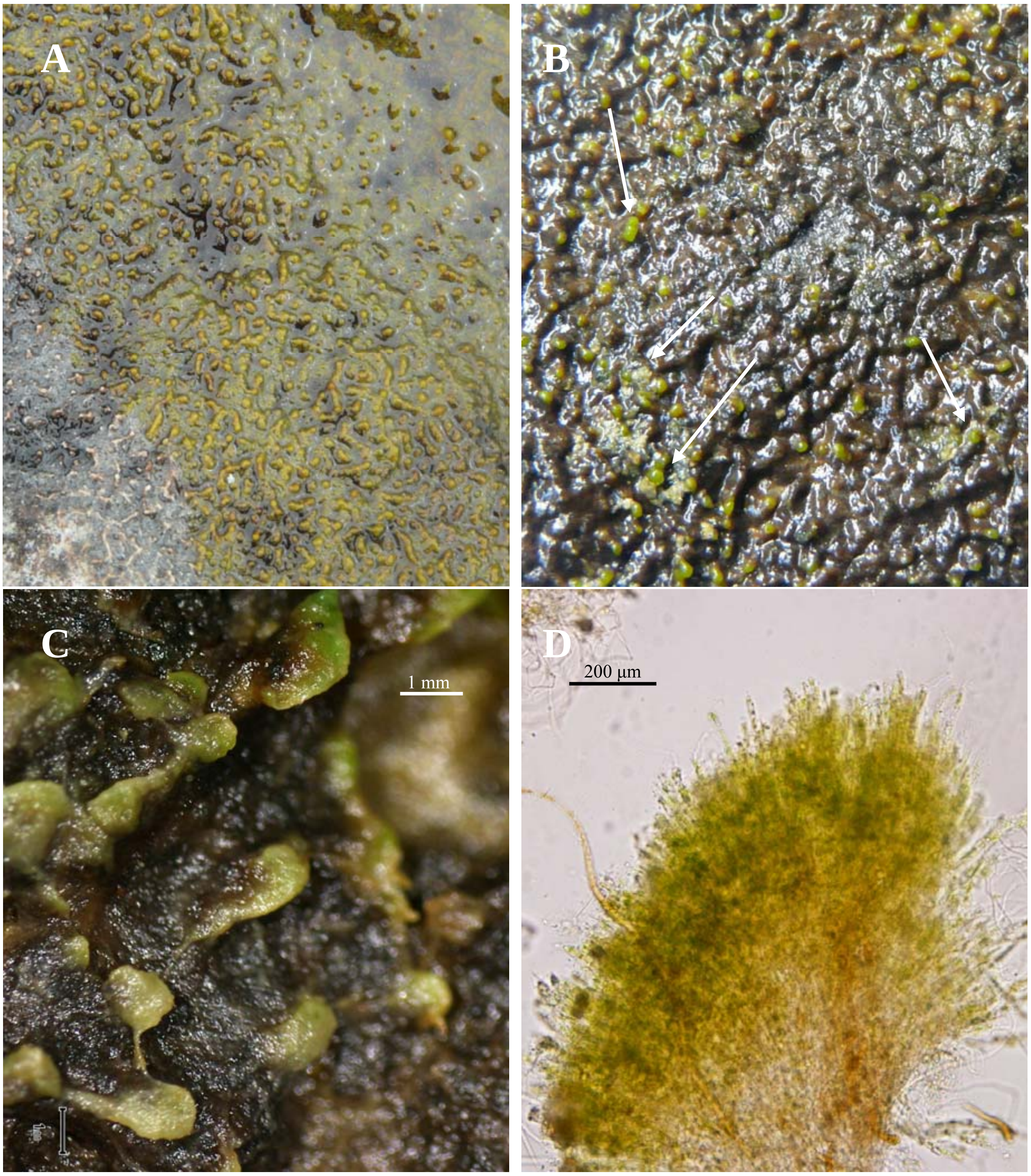
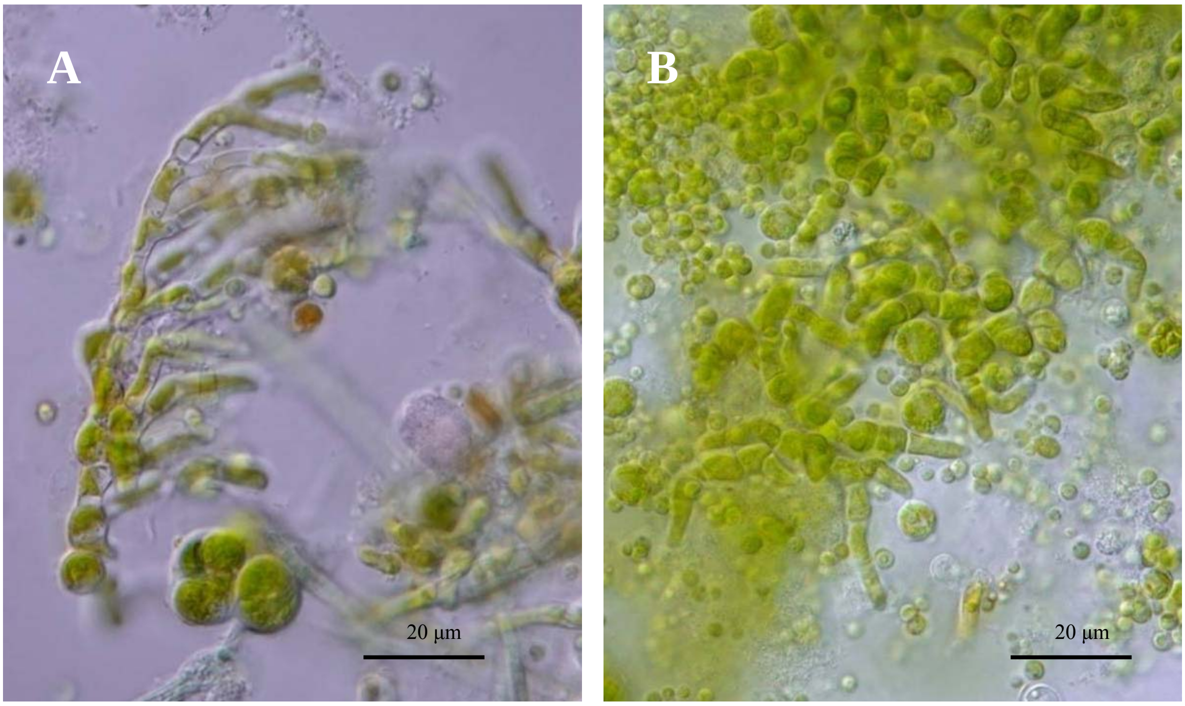


Fig. 7. *Andreassenia antarctica* from cultures.



Invited paper

Specificity of cyanobacterial microflora in Antarctica.

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Introduction:

The cyanobacterial microflora of Antarctica is considered to be mainly secondary. The consequence of such premise was the common practice to identify the Antarctic cyanobacterial populations according to monographs and keys, based mostly on populations from Northern temperate zone (Geitler 1932, Prescott 1951, Starmach 1966 and others), which was also in accordance with the hypothesis of cosmopolitan distribution and very wide ecological plasticity of cyanobacterial taxa.

The easy transport of diaspores by wind and human activities (particularly in the last decades) was supported by the exact studies by Wynn-Williams (1991) and Broady & Smith (1994). The continental input of new cyanobacterial spores to Antarctica surely exists, but cannot explain the special stable diversity of Antarctic cyanobacteria. Our studies indicate the specificity and stability of Antarctic cyanobacterial communities and several modern data derived from recent articles support our results:

Results and Discussion:

1. Recent studies of cyanobacterial flora indicate a high percent of species unknown from other regions. Former authors (West &West 1911, Fritsch 1912, Carlson 1913 and others) described several species from Antarctica which were recently confirmed and are evidently endemic for Antarctica. However, in later period of presupposed "cosmopolitan distribution of cyanobacteria" several floristic papers occurred, in which majority of identified species from Antarctic habitats were designated by names of widely distributed taxa. This happened in spite of the fact, that the taxa were originally described from very different ecological situations (from bark of tropical trees, etc.; see, e.g. in Pankow & al. 1987). From our analyses of cyanobacterial populations, it follows that about 60% of new or not identifiable species are from the King George Island, S Shetland Islands, and only 20-40% of species are known also from regions outside of Antarctica (Komárek 1999). The same situation was found in James Ross Island, NW Weddell Sea, where only maximally 20% of registered morphological taxa have wider distribution and were found earlier outside of continental and/or maritime Antarctica (Komárek & al. 2008). The number of described endemic species from continental Antarctica is already higher than 150, however, many ecotypes still wait for their taxonomic definition.
2. This situation was fully supported by recent molecular analyses of Antarctic cyanobacterial microflora. The relation of published phylogenetic trees to morphological morpho- or ecospecies is usually respected only unsufficiently. Contrastingly, from published and our trees follows, that the Antarctic strains are concentrated mostly in special clusters (Fig. 1) (cf. Nadeau & Castenholz 2000, Gordon & al. 2000, Taton & al. 2003, Jungblut & al. 2005, Strunecký et al. 2009 and others). Of course, the published results do not prove the absence of such clusters from other regions. However, the first results support the high percentage of specific genetically delimited clusters in Antarctic habitats.

3. Recent knowledge of the wide cyanobacterial diversity in relation to special ecological conditions support also the genetic variation among the populations of morphologically similar types. There were found the genetic relations in populations from one extreme habitat (Garcia-Pichel et al. 1998), but also the genetic diversity of morphologically very similar (morphospecies) from geographically and partly ecologically distant localities (Casamatta et al. 2005, Gugger et al. 2005, Strunecký et al. 2009, and others). This process is commonly valid for diversification strategy of cyanobacteria. The stabilization and adaptation of cyanobacterial populations in such extreme conditions, as are Antarctic ecosystems, is evident. This fact coincide to the data about the possible rapid physiological and genetic adaptation of cyanobacterial ecotypes (Hagemann 2002).
4. The ecophysiological adaptations were found in the Antarctic cyanobacterial populations, which proved their long-term adaptations to Antarctic conditions. Rapid utilization of light intensity (measured by chlorophyll fluorescence kinetics – see Fig.2), high rate of special pigments (carotenoids) production in cells of several species, high amount of antioxidants in cells, and the special life strategies enabling the rapid change from vegetative to dormant stages with the ability to survive in dried and frozen state support the idea of long-term adaptation.
5. In numerous Antarctic habitats, such as e.g. lakes, glacial creeks, seepages, wet soils, etc., special complex communities with the same species composition, zonation, succession and proportion of dominants develop repeatedly every year (cf. Komárek & Komárek 2003, Komárek & Komárek 2009, Elster et al. in press,). The dominants are represented by special morphospecies, which are not common in habitats of a region, from which diaspores are transported. The development of the same communities every year proves the existence of stable adapted geno- and morphotypes endemic for Antarctic habitats.

Conclusion:

It is possible to summarize, that Antarctica, as all other continents and regions, is surely open to the transport of cyanobacterial diaspores. Human activities play important role in this process. However, in Antarctica, there exists a wide spectrum of cyanobacterial morpho- and genotypes, which form special communities every year with dominating adapted species, among which numerous Antarctic endemits exist.

Acknowledgements: The authors are indebted to all friends and colleagues, who helped them to participate on the Antarctic expeditions in last years and to study the fascinating world of Antarctic cyanobacteria. The presented conclusions follow from numerous results, obtained under the grant support GA CR 206/05/0253, 206/09/0697, GA AS CR IAA600050704. We appreciate particularly the technical help of D. Švehlová from Botanical Institute of the Czech Academy of Sciences.

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Fig. 1. Phylogenetic tree of cyanobacterial strains according to Taton & al. 2003. The isolated clusters from Antarctica are marked by thick frames and arrows.

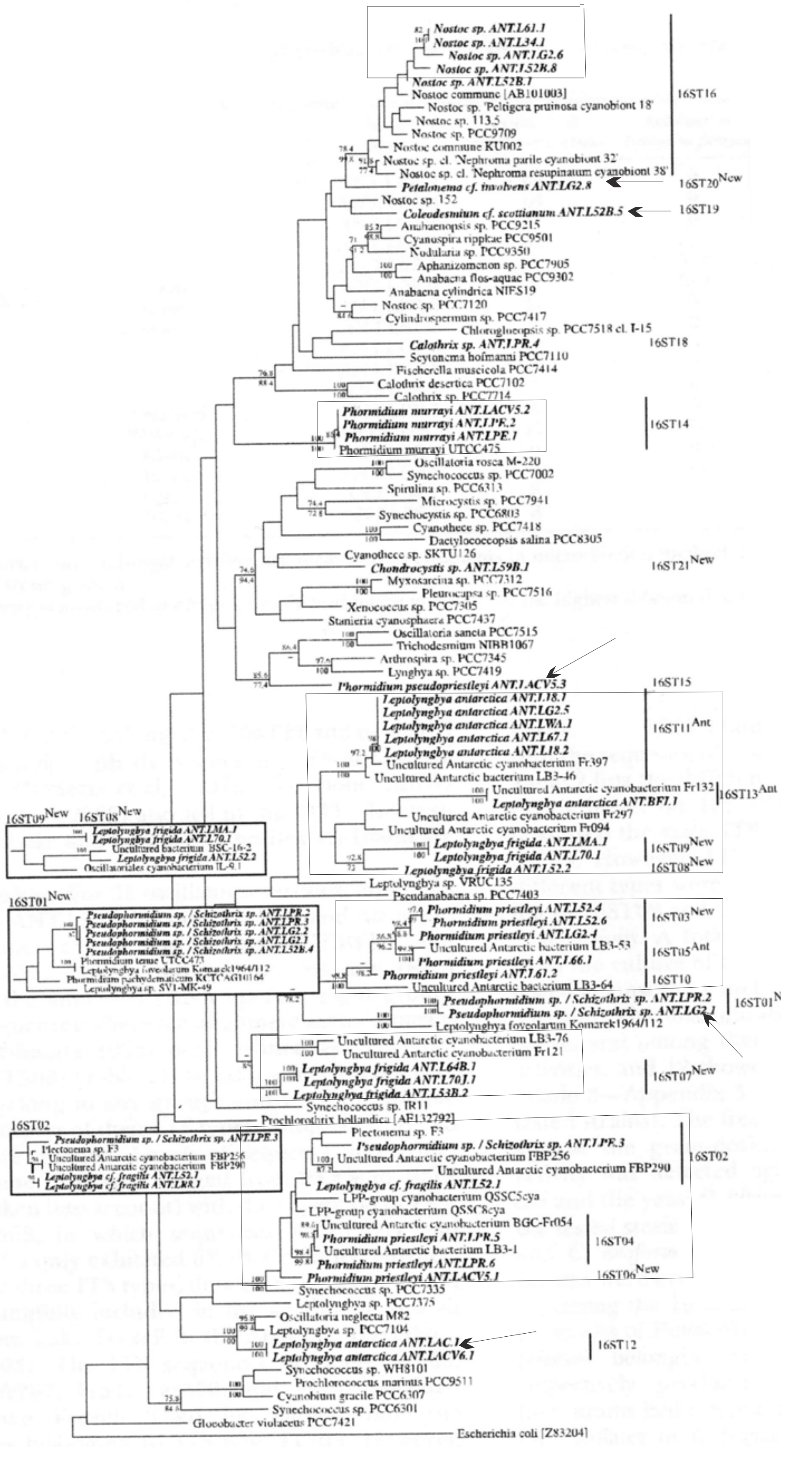
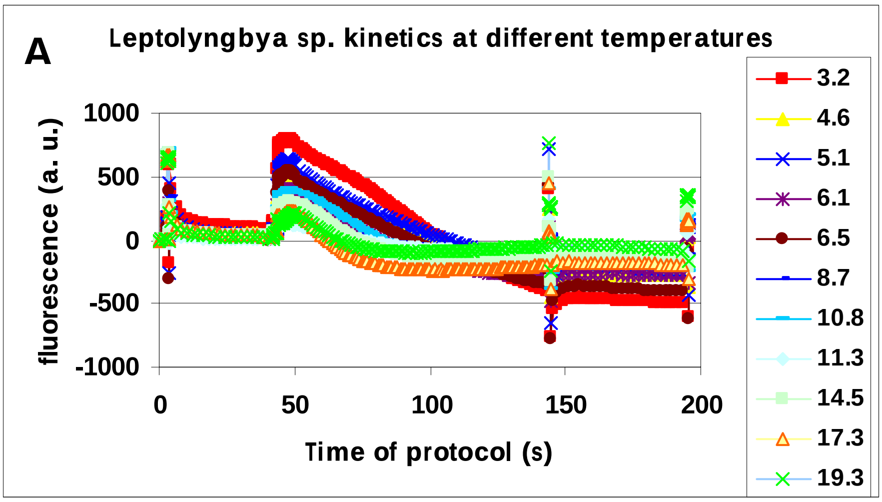
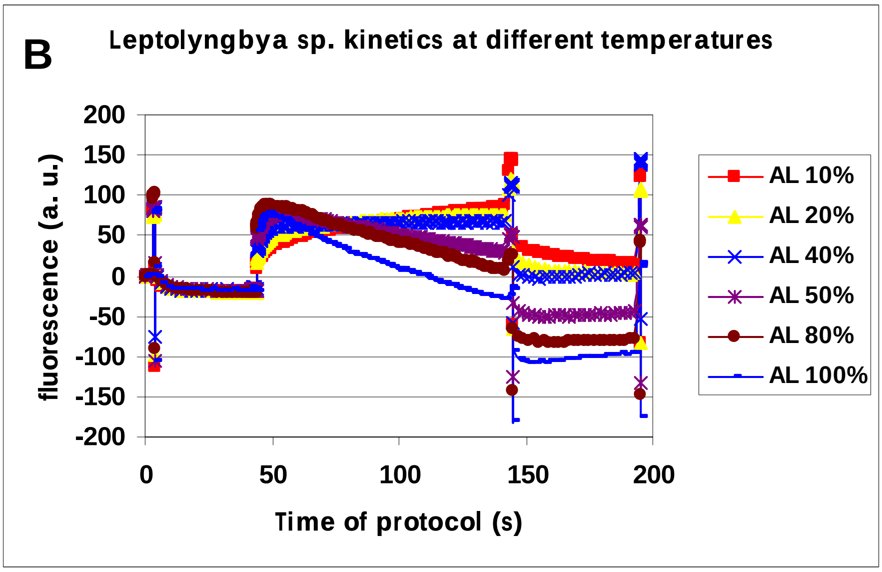


Fig. 2. Induction kinetics of fluorescence in Antarctic *Leptolyngbya* sp. taken under different temperatures (A) and light intensity (B).





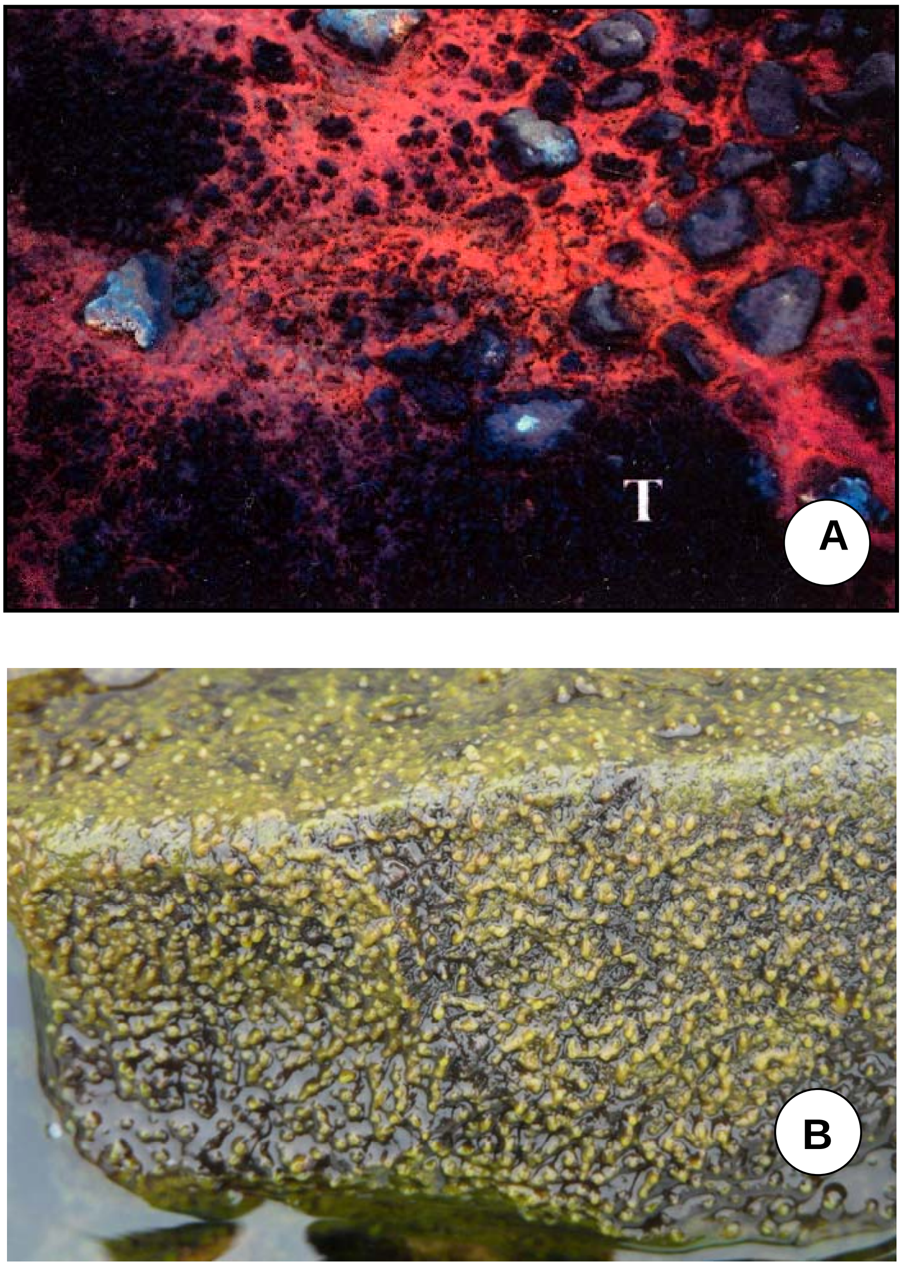


Fig. 3. Special complicated cyanobacterial communities with dominant represented by endemic Antarctic species:

A. Cyanobacterial mats developing every year regularly and with the same zonation and succesion in seepages of maritime and coastal Antarctica. The upper layers are formed by *Leptolyngbya vincentii* (reddish layers) and the end of the vegetation period by the *Nostoc* cf. *commune* and *Tolypothrix* sp. (T).

B. A special type of calcite structures developing in few lakes on James Ross Island formed by a dominant *Calothrix* sp. and subdominant *Coleodesmium* sp., which represent yet the unknown taxonomic cyanobacterial units. (Published with kind permition of J. Elster and L. Nedbalová)

Evaluation of nutrient limitation and heavy metal inhibition in Antarctic streams by a miniaturized algal bioassay

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Introduction:

In the polar regions, shallow freshwater habitats are an information source of the environmental change. It is expected that in these regions, ongoing global changes may affect water properties in future. Chemical weathering processes will likely be more active, and, consequently, the chemical and biological water properties will change.

The miniaturized algal bioassay (microassay) is a simple method for evaluation of water toxicity and water trophy [1]. Two parameters are used in evaluation - the algal growth potential (AGP) and the algal primary productivity rate (APPR). The algal growth potential (AGP) is defined as the highest observed dry weight content reached under optimum cultivation conditions, which reflects the nutrient content of a water sample. The algal primary production rate (APPR) is a growth rate calculated as the slope of a linear regression dependency of the natural logarithm of the dry weight on time, during the exponential phase of the growth curve.

Material and Methods:

The AGP and the APPR of oligotrophic and eutrophic water samples from snow-melt streams (King George Island, South Shetlands, Antarctica) were measured at 5 - 7 °C (AGP_5/$APPR_5$) and 25 °C (AGP_{25}/$APPR_{25}$) in a microassay [1] in order to find possible nutrient limitation and heavy metal inhibition. The other goal was to test the suitability of the microassay for such tests by comparison with the field data. The strain *Sichococcus bacillaris* Elster 1998/28/51, isolated from Arctic soil, was used for cultivation at 5 °C and the temperate one *Stichococcus bacillaris* Hindák 1984/15, isolated from temperate lake Hafnersee, Austria, for cultivation at 25 °C and were obtained from (Culture collection of autotrophic organisms, Institute of Botany, Třeboň, Czech Republic). The water samples were enriched for nutrient limitation tests by 1000 μg.L^{-1} N-NO_3^-, 50 μg.L^{-1} P-PO_4^{3-}, a mixture of N-NO_3^- + P-PO_4^{3-}; for heavy metal inhibition tests by 1000 μg.L^{-1} Na_2-EDTA.

Results and Discussion:

The AGP_5s of the water samples were significantly higher than AGP_{25}. The nutrient limitation tests proved nitrogen deficiency that had been observed *in situ* in oligothophic streams at the King George Island. No nutrient limitation was observed in the mezotrophic stream samples. The heavy metals influence effect on AGP_5 and AGP_{25} was not detected. The microassay proved to be a valuable tool for testing the hypotheses about possible factors affecting the primary productivity in field. Such a conclusion has only limited validity. It is because with respect of limitations given many simplifications in laboratory treatment and experimental design did not allow to study full extent of studied envitonmental factors such as in the field.

Acknowledgements: This research was supported by Project GA AV KJB600050708, and by projects of the Ministry of Education of the Czech Republic, Kontakt ME 934, ME 945, as well as long-term institutional research plan AV0Z60050516.

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Physiological study of Antarctic green algal lichen photobionts: An optimized method of isolation and culturing technique

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Introduction:

Lichens – an unique association of fungus with alga and/or cyanobacteria, are known for their ability to survive extreme conditions of the environment.

In last decade, several authors have focused on study physiological characteristics of lichen photobionts, algae in particular. To characterize physiological properties of algae growing within lichen thalli, it is necessary to separate algal cells from fungal partner by isolation techniques.

Photobiont cells can be more or less successfully isolated from all lichens. Knowledge of lichen anatomy offers important information for isolation of photobionts. Often, it is possible to remove layers of heteromeric thalli type without green photobiont cells mechanically. This can noticeably decrease contamination by cells of mycobiont (Yamamoto et al. 1987). For the isolation, it is suitable to cut lichen thalli into small pieces. Yamamoto et al. (1993) used about 1cm of marginal part of the fruticose thalli or $1cm^2$ of foliose thalli.

Overview of recent techniques:

Lichen material must be mechanically disrupted. For this purpose, a mortar (Yamamoto et al. 1987) as well as an Elvehjen-Potter's homogenizer (Ascaso 1980) is frequently used. Fragments of lichen thalli are homogenized between two slides and single cells are isolated with micropipette by micropipette technique (Ahmadjian 1967a,b, 1993). For a mixed homogenate (with mixed cells of photobiont, mycobiont and contamination), it is recommended to filtrate raw homogenate through several layers of sterile gauze.

From the literature, three basic methods of photobiont selection from the homogenate are known: -1- Yamamoto's method (Yamamoto et al. 1987, 1993) is based on differential filtration of the homogenate through sterile filters with pore size 500µm, followed by filtration through filters with pore size 150µm. Resulting filtrate is washed three times with sterilized water (mineral medium) and transferred on mineral agar medium. -2- Micropipette method is based on direct selection and transfer of cells of the photobiont into sterile water on microscope slide with micropipette (diameter 50 – 75µm) to obtain diluted photobiont suspension. Then, the cells from the suspension are transferred on mineral agar medium. For details see Ahmadjian (1967a, b, 1993) and Ettl & Gärtner (1995). -3- From centrifugation methods is mostly used differential and gradient centrifugation.

In differential centrifugation (Drew & Smith 1967, Orus & Estevez 1984), low-speed centrifugation (<100 x g) is used for removement of big fragment of thalli from the homogenate. For direct collection of the photobiont cells from supernatant, high speed centrifugation (~400 x g) is used. Exact conditions of the centrifugation should be optimized for specific photobiont species. After centrifugation, filtration (membrane filters with pore size 10 – 50µm) of the supernatant can be useful. Then, the filtrate should be washed with sterile water and transferred on cultivation medium.

In gradient centrifugation isolation, disrupted thalli fragments are placed over a density gradient and centrifuged. Separated photobiont cells are collected from particular layer of the gradient. Various chemicals ($CsCl_2$, sucrose, Percoll) are used to prepare density gradient. For detail usage see Ascaso (1980 – $CsCl_2$ sucrose), Calatayud at al. (2001 - Percoll).

The lichen photobiont cells can be cultured on various media (Ahmadjian 1967a, b, 1993). The most common is Bold's basal medium (BBM) (Ahmadjian 1993) and its modifications. Colonies of common photobionts (*e.g. Trebouxia*) cultured on mineral (anorganic) medium can be visible after 2 – 4 weeks from innoculation. BBM medium modified by addition of nitrogen (BBM3N; Ahmadjian 1993) may cause an increase of logarithmic phase of the photobiont growth. Organic modification of BBM medium known as *Trebouxia* medium can be prepared by addition of organic source of nitrogen (peptone, asparagin or kasein) and glucose (Ahmadjian, 1993).

An optimal growth conditions must be determined individually. Generally, photobiont cultures can be cultivated at temperatures from 5 to 25°C, lower irradiance from 50 to 500Lux) and pH from 6 to 7 (Yoshimura et al. 1987, Friedl 1989b, Ahmadjian 1993, Yamamoto et al. 1993).

Optimized Methods:

In our laboratory, we have been dealing with algal partner cultivation for some 6 years. Here we bring an overview of the method used with few optimizing points. For isolation and long-term culturing, two Antarctic lichens (*Umbilicaria antarctica*, *Usnea antarctica*) were used. Lichen thalli were collected at the King George Island, Maritime Antarctica. The thalli were transferred to lab in dry state.

For photobiont isolation from used lichen thalli, we optimize the techniques of differential centrifugation combined with subsequent filtration of cell suspension.

To reach optimal hydration state, the lichen thalli were sprayed with water and placed in Petri dishes in between two sheets of wet filter paper. They were kept at 5°C and weak irradiation ($30\mu mol.m^{-2}.s^{-1}$) for three days. The hydration state and restoration of photosynthetic activity were tested by chlorophyll fluorescence measurement (OptiScience OS-FL1 fluorometer). Optimal hydration state was reached when maximal values of variable fluorescence (F_V) and quantum yield of photochemical reactions in photosystem II (Φ_{PSII}) were reached. Revitalized thalli were rinsed with tap water for 10 min to remove rough dirt and most of potential contaminants. Then, lichen material were cut in small pieces ($>1cm^2$) and homogenized in a mortar with addition of small volume (1-2 ml) of sterile BBM medium. Raw homogenate was filtered through a gauze. Obtained cell homogenate was used in the following steps of photobiont separation by differential centrifugation (Universal 32 R, Hettich Zentrifugen, Tuttlingen, Germany). First, centrifugation at 500 rpm (27 x g) was used for 1 min. to remove big thalli fragments. Supernatant was resuspended and centrifuged at 2000 rpm (440 x g) for 3 min. Obtained sediment was homogenized in sterile BBM medium by vortex (1/2 min.) and centrifuged at 1000 rpm (100 x g) for 1 min. Last step was repeated four times. Then, the cell suspension in sediment was filtered through membrane filter (nylon, pore size 11µm) and washed by sterile BBM medium. The cell innoculum from filter was transferred on agar BBM medium. Photobiont cultures were then cultivated in Petri dishes on agar BBM medium at 5°C and irradiance of $10\mu mol.m^{-2}.s^{-1}$ with photoperiod 16h/8h (light/dark). The above described method brings satisfable results with only limited contamination.

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Lake ecosystems of James Ross Island: short characteristics

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In austral summers of 2008 and 2009, we performed the first limnological survey of lakes on James Ross Island (64°S 58°W, NW Weddell Sea, Antarctica). A total of 34 lakes in the northern deglaciated part of the island were sampled, and their origin, geomorphological and hydrological position, physical, chemical and biological characteristics were examined.

The thermal regime of lakes is mainly driven by their geomorphological and hydrological position. Whereas important diurnal fluctuations were observed in shallow lakes, deep frozen lakes were characterized by very stable temperature conditions (*e.g.* the temperature of Rožmberk lake recorded in 30 min. intervals in January 2009 was 0.5±0,1 °C). Due to bedrock character, pH of lake water was usually neutral to slightly alkaline, the range of conductivity values was 35–3956 µS.cm^{-1} (median: 155 µS.cm^{-1}). Overall, water chemistry was very variable, including nutrient status of particular lakes. Apart from autotrophic assemblages mentioned below, the lake biota was characterized by common occurrence of crustaceans *Branchinecta gaini* Daday (Anostraca) in shallow lakes and *Boeckella poppei* (Calanoida) both in shallow and deep lakes.

Here, we present a short overview of lake types, which can be distinguished on James Ross Island, they are summarized descendently according to their origin and geomorphological and hydrological stability:

(i) Stable shallow lakes of higher-lying levelled surfaces. These are the oldest lakes in the studied region; last deglaciation of these glacially-levelled volcanic surfaces took place during the Termination I and early Holocene, some 7–12 ka BP and they may originate after that. Their winter cover melts at the beginning of summer. A very specific microflora develops especially in seepage lakes.

(ii) Shallow coastal lakes. These lakes developed mostly in the mid Holocene, after the marine and/or glacier retreat of the coastal areas. The well-developed littoral assemblages occur in these very shallow lakes (max. depth ~0.2 m). However, their growth is limited by high aeolian input of silty to sandy material originating mostly from cretaceous sediment; their bottoms are therefore formed by thick layer of sediments.

(iii) Stable lakes in old moraines. Advancing local glaciers accumulated large moraine ridges during the Holocene, after their decays numerous lakes developed. Only the largest and most stable outlast until present. Similarly to previous types, benthic mats are formed by a characteristic microflora with cyanobacterial morphospecies known from continually frozen lakes in continental Antarctica.

(iv) Small unstable lakes in young moraines. Young moraines in front of retreating local glaciers host small lakes, they mostly have stony bottoms, where rich cyanobacterial assemblages often develop in form of structured mats up to several cm thick.

(v) Stable deep cirque lakes. Deep glacier cirques developed on the lee sides of volcanic mesas, where accumulation of drifting snow allowed cirque glacier formation. Due to recent decay of local glaciers at the James Ross Island numerous cirque lakes evolved. Their key feature is the development of extensive ice cover, which persists at least partly during summer. The ice cover can be up to 2 m thick and has a characteristic structure composed of long crystals (“candle ice”). Littoral assemblages are poorly developed and primary producers are mainly represented by phytoplankton.

(vi) Thermokarst lakes. The youngest and less stable lakes are thermokarst lakes, which evolved in debris covered glacier systems. These lakes can be quite deep (> 10 m), and represent the most abundant lake type on James Ross Island. Unstable stone cover of steep icy shores does not sustain the development of microbial mats.

It can be concluded that lakes on James Ross Island represent a unique set of diverse ecosystems in the transitional zone between maritime and continental Antarctica, thus playing an important role in comparisons with other lake districts in Antarctic region (*e.g.* Livingstone Island, McMurdo Dry Valleys, Amery Oasis, Bunger Hills).

Variability of oxygen content in different lakes of the James Ross Island as dependent on actual weather conditions

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Introduction:

Amount of dissolved oxygen is considered both biological and physical factors limiting life and development of organisms in Antarctic lakes. In those lakes with substantial amount of autotrophic organisms and partial ice cover, water might be supersaturated by oxygen [1]. Actual amount of dissolved oxygen depends on physical and chemical characteristics of water of particular lake as well as the development of biota [2]. All these factors may cause variability of dissolved oxygen within a single austral summer season.

At the James Ross Island, there is a long-term research of lakes performed by Czech ecologists, limnologists and botanists (see *e.g.* [3], [4], and the abstracts and papers of Nedbalová, Elster and Komárek in this volume). Majority of the lakes at the Northern part of the James Ross Island has been already investigated by the above team and oxygen concentration measured in order to evaluate overall lake character. In this paper, we present some additional information on variation in oxygen concentration of particular lakes. Particularly, we focus on differences related to actual weather conditions. Here we present a comparative study of dissolved oxygen concentration in different lakes masured at typical sunny and cloudy days during austral summer.

Sampling site	Lake code No.	Lake provisional name	Latitude	Longitude
1	DULAN6	Dulánek	-63,8170399	-57,8458549
2	LACH1	Interlagos 1	-63,7985567	-57,81012
3	LACH2	Interlagos 2	-63,7989033	-57,8104816
4	LACH3	Lachman 2	-63,7998683	-57,8087217
5	LEDN4	Ledňáček	-63,8097449	-57,8239266
6	NADEJE5	Naděje	-63,8140132	-57,833795
7	ROZMB7	Rožmberk	-63,817925	-57,844955

Table 1. Overview of the lakes and sampling sites included into comparative oxygen measurements.

Material and methods:

Concentration of dissolved oxygen were measured in seven selected lakes of different age and origin (for nomenclature, see the Abstract by Nedbalová et al. in this volume) in Northern part of the James Ross Island. Geographic location of the lakes under investigation is shown in Table 1. The lakes abbreviated as LACH1, LACH2, and LACH3 are located in Eastern part of Lachman cape, 18 m a. s. l. There are no neghbouring rocks or high walls, therefore these localities are exposed to sunlight during majority of a day. The depth maximum of LACH1, LACH2, and LACH3 vary in the range from 0.5 to 1.5 m. More detailed description of the lake (here referred as LACH3) is given in the contributed paper by Komárek et al. in this volume.

The LEDN4 lake is located at the foot of the Berry Hill messa at the height of about 180 m a.s.l. It is a typical thermokarst lake which exhibited a rapid decrease in water level within several weeks during austral summer of 2008. The NADEJE5 is a large lake located in between two rocks forming Southern walls of the Berry Hill. The lake is located at about 220m a.s.l. Almost permanent shady conditions are presumably a cause of permanent ice body inside the lake. The DULAN6 lake is located at the foot of the Windy pass with eastern exposition, about 220m a.s.l. It is a small (~20m^2) reservoir in a shallow depression. The ROZMB7 lake is of silimar type and general characteristics as the NADEJE5 lake. It is located under the northern hillside of the Lachman crags, about 200m a.s.l. With direct sunlight during lasting for half of a day, there is relatively smaller ice body inside the lake.

On January 12th (sunny day) and January 20th 2009 (cloudy day), all the sampling sites (see Table 1) were visited and records of oxygen concentration taken. The oxygen concentrations were measured by an Oxymeter – WTW Oxi 197i equiped with a WTW CellOx 325 electrode. The probe was placed into the depth of about 20 cm and oxygen concentration was measured after reaching equilibrium and constant value. At least 15 replicates were measured at each sampling site.

Results and Discussion:

In all cases, dissolved oxygen concentrations were found lower during the cloudy day than the sunny day (see Fig. 1). As expected, maximum values were found in coastal lakes with clear water at low altitudes at the sunny day. Such finding might be attributed to rich autotrophic microflora, and, especially, high water temperature that was reached in LACH1, LACH2 sites thanks to full sunshine reaching the lakes and their shallow water profiles. This might be well documented for LACH1 (13.6 ^{o}C) when compared to the shaded and higher altitude-located NADEJE5 (1.0 ^{o}C). Lowest oxygen concentration was recorded in the ROZMB7 sampling site, at which oxygen concentration was by one third lower than maximum found in the LACH1. This was caused by ice cover that was disintegrated only few meters along lake margins while vast majority of the lake (ROZMB7) was covered by ice in the half of January. Also low water temperature might interact. However, even in the ice-covered lake, statistically significant difference in oxygen concentrations was found between sunny and cloudy (overcast) day.

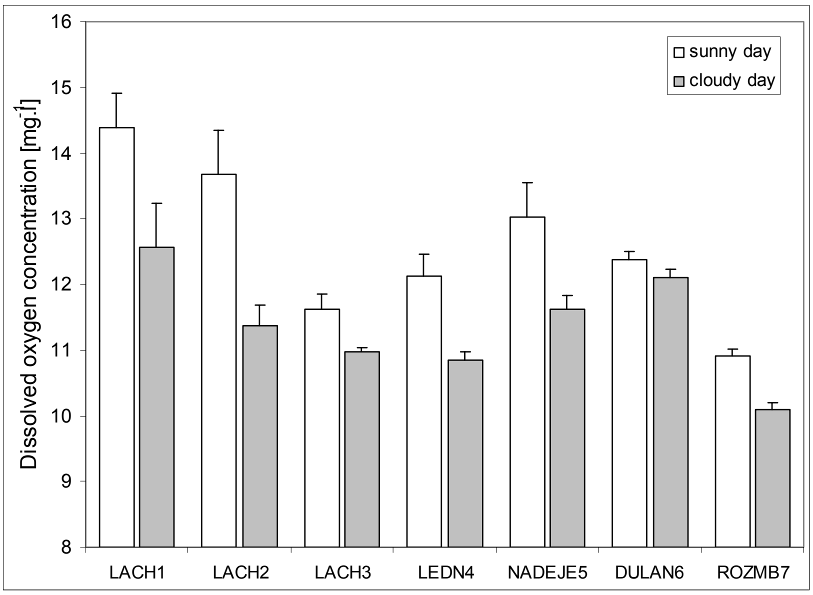

Fig. 1. Oxygen concentrations in several lakes at the Northern part of the James Ross Island recorded for typically sunny and cloudy day, respectively (January 2009).

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Biogenic calcite structures in Green Lake, James Ross Island, Antarctica

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Introduction:

Microbial carbonates (stromatolites, thrombolites and other forms of calcified and spar-encrusted microbes) have been important structures since the Archean. Their formation is biologically-stimulated, however, it also requires a favourable pH, calcium concentration and other ecological conditions. The associated microbes (microscopic organisms) include bacteria (especially cyanobacteria), fungi, micro-algae and protists. Cyanobacteria and micro-algae play a key role in microbial carbonate formation. The main processes of microbial carbonate formation include grain trapping and calcium carbonate precipitation. Cyanobacteria and micro-algae thrive in shallow-water oxygenated environments in the water column and near the sediment-water interface, mainly in extreme environment. As a part of limnological study of the northern part of James Ross Island, Antarctica, we found two lakes, where unusual periphyton communities. These communities trap grain and precipitate calcium carbonate on their cells surfaces. These communities can be classified as thrombolites.

Lake description:

In north-east deglaciated part of James Ross Island, Antarctica, we studied two shallow lakes located between Andreassen point and Solorina Valley. They were Green Lake I and Green lake II (Fig. 1 and 2), in which a special cyanobacteria-microalgae communities (thrombolites) have developed. General characteristics of the two lakes are as follows. Both lakes are shallow (up to 180 cm) with surface area about 1000 to 2000 m^2. Both lakes fluctuate in their water level, because they have inflow from snow-fields only during snow melt period. Water temperature during summer can reach 10°C in both lakes, water is saturated by oxygen with neutral or slightly alkaline pH. Water is extremely poor in nitrogen content (NO_2-N, NO_3-N, NH_4-N, TN). On the contrary, the water is relatively rich in phosphorus (TP, TDP, SRP) and calcium content. Cyanobacteria-microalgae thrombolite communities were found across both lakes from dry littoral (at summer time) to the deepest central part of lakes.

Fig. 1.

Fig. 2.

Thrombolite description:

Thrombolite community (Fig. 3) is composed of two major parts: 1) Cyanobacterial black biofilm (Fig. 4) that is formed mainly by *Calothrix* sp. and, to a lesser extent, simple filamentous *Phormidium* sp. *Leptolyngbya* sp. and others, 2) Micro-algal part is predominantly composed of unknown green branched filamentous algae (Fig. 5) from Chaetophorales group. At present, the intensive polyphasic study of both dominant species is under progress. Visible calcium carbonate crystals (Fig. 6) together with mineralized biofilm (Fig. 7) have been observed.

Fig. 3.

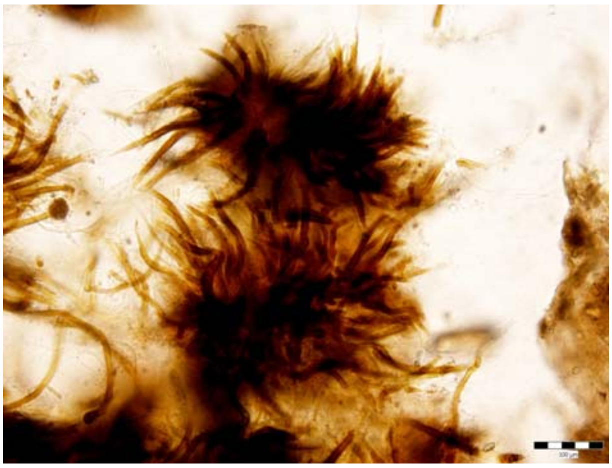

Fig. 4.

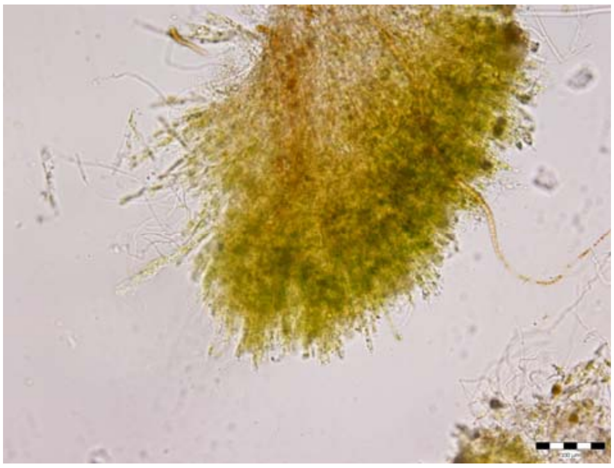

Fig. 5.

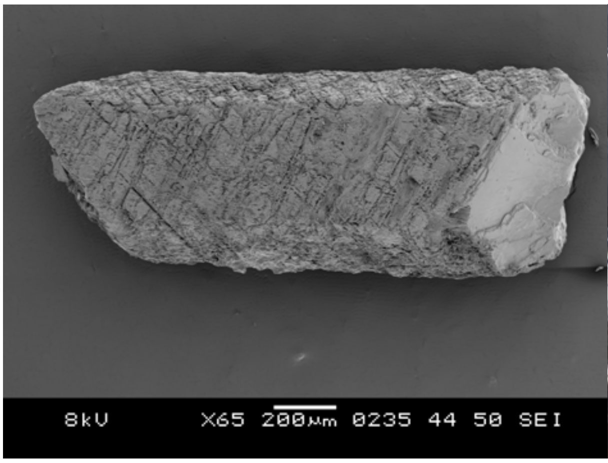

Fig. 6.

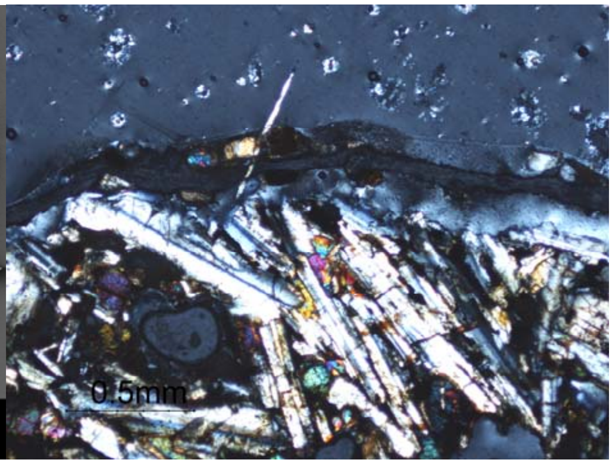

Fig. 7.

Conclusion:

This is the first record of cyanobacteria-microalgae thrombolite community in the Antarctica. At present, an intensive study of this community is performed. The study will answer to the following questions:

1. What is the age of this community?
2. Which inorganic substances are precipitated by microbial lithogenetic processes?
3. What is the origin of species which compose the community?
4. What ecological conditions the microbial community requires for its development?

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Overview of studies on utilization of vascular plants by Antarctic birds at nesting sites

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Introduction:

During the last 50 years, the Maritime Antarctic experienced significant climate warming. As a consequence, population of the only two vascular plants native to the Antarctic, i.e. *Deschampsia antarctica* and *Colobanthus quitensis* dispersed and established over previously unoccupied territories. There are different hypotheses about the time span of their initial invasion of this region. Being successful, both species must be effectively adapted to spread over ice-free areas isolated by vast seas and/or glaciers. Their seeds may have been imported by wind and/or by birds. While winds most probably disperse only larger propagules within habitats, birds may carry also tufts probably not only among the (sub-)Antarctic islands but also from South America. There is a number of indications that birds can effectively disperse vegetative plant material. Smaller or larger vegetative parts of the Antarctic vascular plants are able to survive transportation by birds and can successfully established in suitable environmental conditions in yet uncolonized localities. However, the bird species involved, the manner(s) of plants' engagement, and itineraries of their travels remain unclear. Therefore, the aim of the present study was to investigate utilization of vascular plants in the nests of *Larus dominicanus* and both *Catharacta* species at two distant locations in the Maritime Antarctic.

Materials and Methods:

The field survey was conducted during the austral summer seasons 2006/07 and 2007/08. It involved investigations of nesting habits of *Larus dominicanus* and two skua species, *Catharacta maccormiki* and *Catharacta lonnbergi*, at two distant locations, i.e.: the Point Thomas area (King George Island) and the Argentine Islands region including a few sites on the western shore of the Antarctic Peninsula. All accessible nests of the investigated bird species were inspected for presence of parts or whole vascular plants. On average 20 nests of each bird species were investigated in each region. All the investigated nests were photographed. To avoid disturbance to occupied nest their inspections were performed with special care and in short sessions only.

Results and Discussion:

The data provide evidence that the investigated birds significantly contribute to dispersal and establishment of the Antarctic vascular plants. The results also demonstrate that *L. dominicanus* at both investigated regions use vascular plants for nest building on a regular basis. Besides dried fragments of both vascular plants species, alive and well rooted clumps were also observed in the gulls' nests. The utilization of vascular plants by gulls is, therefore, independent of the geographic location. In case of skuas' nests (the species were not distinguished), vascular plants were found in all nests from the Point Thomas area, while the nests from the Argentine Islands region contained almost exclusively mosses. It seems that only *C. maccormiki* is present on the Argentine Islands region, while in the Point Thomas area both species, *C. maccormiki* and morphologically similar but larger *C. a.lonnbergi*, occur. The use of vascular plants in nests building seems to be species-specific. However, both species and their hybrids use only plants from immediately adjacent territories and both mosses and vascular plant appear to be utilized by these birds. Therefore, the species-specificity in the use of vascular plants may arise only from plant communities surroundings their nesting sites. Therefore it seems that *L. dominicanus* is probably an important vector distributing vascular plants over the Maritime Antarctic. The distance of such transport needs more detailed investigation. *Catharacta* species, on the other hand, are probably responsible for local propagation, mixing or rejuvenation of the populations.

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Comparative study on utilization of vascular plants by Antarctic birds

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Key-words: *vascular plants, birds, maritime Antarctic*

Introduction:

During the last 50 years, the region of Antarctic Peninsula and adjacent archipelagos, also known as the Maritime Antarctic, experienced significant climate warming. As a consequence, populations of the only two vascular plants native to the Antarctic, i.e. the Antarctic hairgrass *Deschampsia antarctica* Desv. and the Antarctic pearlwort *Colobanthus quitensis* (Kunth.) Bartle. dispersed and established over previously unoccupied territories [1, 2, 3]. There are different hypotheses about the time span of their initial invasion of the Maritime Antarctic region. It may well have happened either after Pleistocene glaciations [4, 5] or even before the maximal ice sheet formation [6]. Being successful, both vascular plant species must be effectively adapted to spread over ice-free areas isolated by vast seas and/or glaciers. Their seeds may have been imported by wind or by birds. However, it has been recorded that the quantity of dispersed seeds rapidly decreases with distance [7]. While winds most probably disperse only larger propagules within habitats, birds may carry also tufts and they can disperse them not only among the (sub-)Antarctic islands but also from South America. There is a number of indications that birds can effectively disperse vegetative plant parts [4, 8]. Smaller or larger vegetative parts of Antarctic vascular plants are able to survive a several days-lasting transportation by birds and can, therefore, successfully established in suitable environmental conditions in yet uncolonized locations [9]. However, the bird species involved, the manner(s) of plants' engagement, and itineraries of their travels remain unclear. This information is very important for investigation of the history and modern aspects of the biology of both native Antarctic vascular plants. This implies a question: What species of birds could have been the carriers of *D. antarctica* and *C. quitensis* during their initial colonization and later dispersal? For example, small migratory birds of the genus *Muscisaxicola* (Tyrannidae), which are common on the bogs where *C. quitensis* thrives in the Andes, have been put forth as an exotic candidate carrier of the Antarctic vascular plants from South America to the maritime Antarctic [10]. Based on the literature data, it seems likely that only three species of the Antarctic birds are probably capable of distributing the vascular plants, i.e.: the Kelp Gull (*Larus dominicanus*), and two skua species the South Polar Skua (*Catharacta maccormiki*), and the Brown Skua (*Catharacta lonnbergi*) [11]. In addition, Southern Giant Petrel (*Macronectes giganteus*) has also been reported to use the grass as nesting material [12, 13]. However, this species is known to be very vulnerable to human disturbance. There are evidence that one breeding colony of this species from the Point Thomas area (King George Island) was completely abandoned due to human disturbance [14]. Therefore, to avoid any disturbance to occupied nests we did not included this species into our investigations. Potentially, all these bird species are long-distance migrants between South America and Antarctica, or even within a wider range [13, 15]. The aim of the present study was to investigate utilization of vascular plants in the nests of *L. dominicanus* and both *Catharacta* species at two distant locations in the Maritime Antarctic.

Materials and Methods:

The field survey was conducted during the austral summer seasons 2006/07 and 2007/08 within the 11th and 12th Ukrainian Antarctic expeditions. We involved investigations of nesting habits of the Kelp Gull (*Larus dominicanus*) and two skua species, the South Polar Skua (*Catharacta maccormiki*) and the Brown Skua (*Catharacta lonnbergi*) at two distant locations, i.e.: the Point Thomas area (Admiralty Bay, King George Island, South Shetland Islands) and the Argentine Islands region. The latter included some of the Argentine Islands: Galindez (where Faraday/Vernadsky Station is located), Skua Island, and several islands of the nearby archipelago: Berthellot Is., Petermann Is. and Yalour Is. as well as a few sites on the western shore of the Antarctic Peninsula. All accessible nests of *L. dominicanus, C. maccormiki*, and *C. a. lonnbergi* were inspected for presence of parts or whole vascular plants. On average, 20 nests of each bird species were investigated in each region. However, the number of nests inspected was limited by the size of the local breeding populations. For example in case of the Kelp Gull (*L. dominicanus*) in the Argentine Islands region, a total of 35–40 nesting pairs have been reported, but only 3–5 pairs are known from the Galindez Island [13]. Moreover, nests of this species are often difficult to reach. Hence, the real number of accessible nests was only about ten [Chesalin, Dyyky personal communications]. In the Point Thomas area, the number of breeding pairs is also relatively low and amounts 13–22 pairs [14, Trivelpiece W. & Trivelpiece S., personal communication]. In case of the South Polar Skua (*C. maccormiki*), approximately 100 pairs nest in the Argentine Islands region, 21–37 of which nest on Galinez Island [13; Dyyky personal communications]. *C. maccormiki* is also fund in the Point Thomas area where its population was estimated for 19 pairs [13, Trivelpiece W. & Trivelpiece S., personal communication]. On the other hand, breeding pairs of the Brown Skua (*C. a. lonnbergi*) occurs only the Point Thomas area, where its population was estimated at 37 breeding pairs [Trivelpiece W. & Trivelpiece S., personal communication]. To avoid disturbances to occupied nests their inspections were performed with a special care and in short sessions only. All the investigated nests were photographed. In addition, photographs of *L. dominicanus* nests (kindly provided by Malgorzata Korczak and Anna Gasek) from the Point Thomas area taken during the summer season 2008/09 were analyzed. Furthermore, 10 photographs of *C. maccormiki* nests published Peklo [13] were also included into the analysis.

Results and discussion:

The data provide evidence that the investigated birds contribute significantly to dispersal and establishment of the Antarctic vascular plants. The results also demonstrate significant differences in utilization of vascular plants between the Kelp Gull (*Larus dominicanus*) and both skua species (*Catharacta* spp.). *L. dominicanus* at both investigated regions use vascular plants for nest building on a regular basis. Besides dried fragments of both vascular plant species, alive and well rooted clumps were also observed in the gulls' nests. The utilization of plants by the gulls, therefore, seems to be independent of the geographic location. In favorable weather conditions, breeding season of *L. dominicanus* starts already at the beginning of October [13]. The gulls mate, defend their breeding territories and collect material for their nests when most of their breeding territories and the surrounding areas are still covered with snow. Facing shortage of a nest material, they might be forced to collect it from distant locations, even from other islands. Thus they may contribute to dispersal and establishment of plants in new locations. In addition, gulls significantly fertilize the soil around their breeding territory, both with their droppings and food remains (*e.g.* shells from cracked limpets). It is well known that, although *D. antarctica* has a wide ecological amplitude, the most favorable conditions for its growth occur in moderately manured areas near bird breeding sites [3, 16]. Therefore, after the introduction of

D. antarctica and *C. quitensis*, brought with the nesting material, development of the Antarctic herb tundra formation becomes possible. Nesting sites may then be abandoned and overgrown with the grass, allowing for development and establishment of the vascular plant communities in a new location. Therefore, *L. dominicanus* seems to be particularly important vector transferring seeds and other propagules among the islands. In case of skua, vascular plants were also found to be important component of their nests. In the Point Thomas area, some vascular plants were found in all the investigated skuas' nests (the species have not yet beed distinguished). On the other hand, skuas' nests from the Argentine Islands region contained almost exclusively mosses. It seems that only *C. maccormiki* is present on the Argentine Islands region, while in the Point Thomas area both *Catharacta* species occur, (*C. maccormiki* and morphologically similar but larger *C. a. lonnbergi*). Although all vascular plants, mosses and lichens are utilized by these birds, the use of vascular plants in nests building appears to be species-specific. However, the differences may result from the fact that both species and their hybrids use only plants collected directly within their breeding territories and their immediate surroundings [Trivelpiece W. & Trivelpiece S., personal communication]. At the beginning of their breeding season, most of the ground is already snow-free. Thus, it is not necessary for these birds to search for nesting material away from their breeding sites. Therefore, the species-specificity in the use of vascular plants may arise only from the plant communities available in the neigbourhood of their nesting sites. On the Barton Peninsula (King George Island), nests of *C. a. lonnbergi* are situated closer to the shoreline and at lower elevation than those of *C. maccormiki* (mixed pairs also tend to occupy lower sites) [17]. Observations from the Fildes Peninsula (King George Island) [15] also support that view. *C. maccormiki* use more often lichens (*Usnea sp.*) to build their nests, but *C. a. lonnbergi* seems to prefer mosses as a nests material. This depends on the surroundings, if it is wet or dry, as well as altitude and type of vegetation present in their breeding territories. The bigger and stronger *C. a. lonnbergi* usually occupies territories closer to penguin colonies, and partly in lower, moister areas where mosses are more abundant. *C. maccormiki* is smaller than *C. a. lonnbergi*, and therefore seems to be forced to use the remaining areas, such as locations at higher elevations where plant communities are usually dominated by lichens [Peter, personal communication]. In the Point Thomas area, both species of skua occur. The Antarctic herb tundra formation is well-developed and occupies significant area here. As a consequence, vascular plants are found in nests of both species. The birds preference to collect plants (as a nest material) found only within their breeding territories is also evident from the observations done at Barton and Fildes Peninsula. Although at both these locations both species of vascular plants occur, they were not found in skuas' nests [15, 17]. Such a finding might be explained by location of nests and their distance from vascular plant communities. The nests of *C. maccormiki* may include vascular plants, though such cases are rare, as this species usually nests in sites located on higher elevation where vegetation is dominated by lichen and moss communities (both at the Point Thomas area and on the Argentine Islands), while most sites with such plant communities in the Argentine Islands region occur at low elevation close to the sea-shore. Therefore, the skuas nesting there seem to contribute only to local rejuvenation and mixing of clumps but not to long-distance dispersal which is probably accomplished by other means. All these observations indicate differences in the use of vascular plants in nest building between *L. Dominicanus* and *Catharacta* species. *L. dominicanus* seems to collect selectively tufts of *D. antarctica* among which, parts of *C. quitensis* might be sometimes found. On the other hand, *Catharacta* species are probably not selective in choosing nest building material. They probably use only plant material available in close vicinity of their nest. Thus, the composition of the plant material in their nests corresponds to the composition of the nearby plant communities. Differences in nest material preferences between *C. a. lonnbergi* and *C. maccormiki* might be explained solely on locations of their breeding territories. In spite of the fact that a series of reports on the nature of the investigated subject are available [17, Peter, personal communication], the issue requires further investigation. Among the investigated bird species, *L. dominicanus* seems to be the most important vector distributing vascular plants and/or their parts over the maritime Antarctic. The distance of such transport needs more detailed investigation. Both *Catharacta* species, on the other hand, are probably responsible only for local propagation, mixing or rejuvenation of the vascular plants populations.

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Long-term study on vegetation responses to manipulated warming using open top chambers installed in three contrasting Antarctic habitats

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Introduction:

There are several reports that Antarctic terrestrial ecosystems located along the Antarctic Peninsula have experienced local warming within last few decades [1, 2]. Both short and long-term responses to warming of the terrestrial ecosystems are observed and reported as e.g. increased rate of deglaciation [3]. Therefore, the attention of polar researchers is attracted to study the likely responses of vegetation to increased temperature in the region. Among several approches, the following have been used in polar regions to increase air/soil temperature on long-term basis: (1) FATI system was used in Greenland on grassland tundra ecosystem [4, 5]. (2) Open top chambers (OTCs) have been used more frequently both in the Arctic, (ITEX tundra project [6], Svalbard [7]. In Antarctica, OTCs have yet been used only in a few locations: e.g. Taylor Valley (USA program), Yukidori station (Japanese program), and along a longitudal gradient consisting of Falkland Is, South Orkneys and Leoni Island [8]. The Dutch Program studies the effect of warming on soil biological activity and biodiversity (for details see e.g. TARANTELLA web page). In this paper, we bring an overview of Czech long-term program aimed to monitoring of changes induced by manipulated environmental warming at the James Ross Island, Antarctica. In this paper, descriprion of the experimental approaches comprising a) OTCs constructions and location, b) OTC-induced microclimate alternation, and c) biophysical measurements of physiological state of Antarctic lichens, mosses is given.

Material and Methods:

Open top chambers microclimate

Within the years of 2007-2009, nine OTCs were established in the Northern part of the James Ross Island in a close vicinity of the Czech Antarctic station Johann Gregor Mendel (63° 50´ S, 57° 50´ W). Hexagonal OTCs were made of 6 plates of a 5 mm thick extruded PMMA plexi glass. An individual plate was of trapezoid shape having the base/top/height dimensions of 70/50/50 cm. The contact line between two neighbouring plates was reinforced by an aluminium belts and fixed by 6 screws each. The whole OTC construction was placed at an experimental plot and fixed to the surface by iron ropes fitted to the top parts of the OTCs.

Location name	Location number	Altitude (m a.s.l.)	Number of OTCs	Site description
Vicinity of the J.G.Mendel station (seashore)	I	8 m	3	Coastal vegetation oasis
Berry Hill mesa	II	350 m	3	Table mountain, basaltic rock
Davies Dome	III	410 m	3	Ice-free glacier margin

The OTCs were installed over typical vegetation cover: (I) moss carpet with several individual spots of lichen growing on soil and stone surfaces, (II) lichens *Usnea antarctica* and *Umbilicaria decussata* located on basaltic stones forming a polygonal structure, and (III) inicial stages of vegetation succession forming algal biofilms and tiny spots of microlichens on deglaciated stony substrates. Temperature sensors (Cu/Co) were installed and connected to a multichannel datalogger (Minicube VV/VX, EMS, Czech Republic). The sensors were put (*a*) into the height of 30 cm above surface, into the substrate to the depths of (*b*) 5, (*c*) 10, (*d*) 15 cm and also (*e*) into the surface cover (moss or lichen). The temperature data are taken in a 30 min step throughout whole year. Additionaly, automatic weather station (Modulog, EMS, Czech Republic) data recorded in the same interval are used to compare the OTC microclima with reference climate (see contribution of Láska and Prošek in this volume).

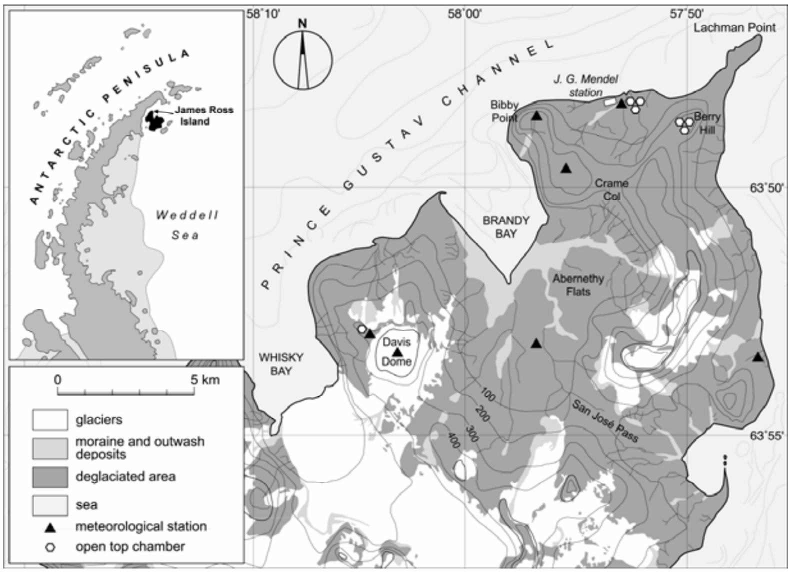

Fig. 1. Three locations of open top chambers included to this study (James Ross Island)

Chlorophyll fluorescence measurements

To evaluate the effect of altered microclimate inside an OTC on photosynthetic processes of lichens and mosses, several methods and instruments were used. In austral summer 2008, a modified PAM-210 (H. Walz, Germany) was installed into an OTC to monitor effective quantum yield of photosynthetic processes in photosystem II (Φ_{PSII}) in *Bryum* sp. in 15 min. interval for 21 d. In such a way, diurnals of Φ_{PSII} were obtained indicating particular periods of photosynthesis enhancement and/or limitation. In January-February of 2009, paralel measurements with two fluorometric systems were performed. The first system was a multichannel monitoring fluorometer Moni-PAM (Heinz Walz, Germany). In our field experiment, we used 2 monitoring heads, one located above moss cusion inside an OTC, the other one at the control plot. We measured Φ_{PSII} in light-adapted state under natural irradiantion each 15 min for 53 days (Barták et al. unpublished data). Simultaneously, measured photosynthetically active radiation (PAR) enabled the calculation of photosynthetic relative electron transport rate $ETR = \Phi_{PSII} * PAR * 0{,}84 * 0.5$ and construction of diurnals for Φ_{PSII} and ETR. The second system consisted of two FluorPen fluorometers (a modified FP-100, Photon Systems Instruments, Czech Republic) equiped with a special power unit. The FluorPen fluorometers were installed in January 2009 and since that time they have been measuring photosynthetic Φ_{PSII} in 30 min interval. Fluorometric data obtained from all the three systems are analyzed and will be published in forthcoming paper (manuscript in prep.)

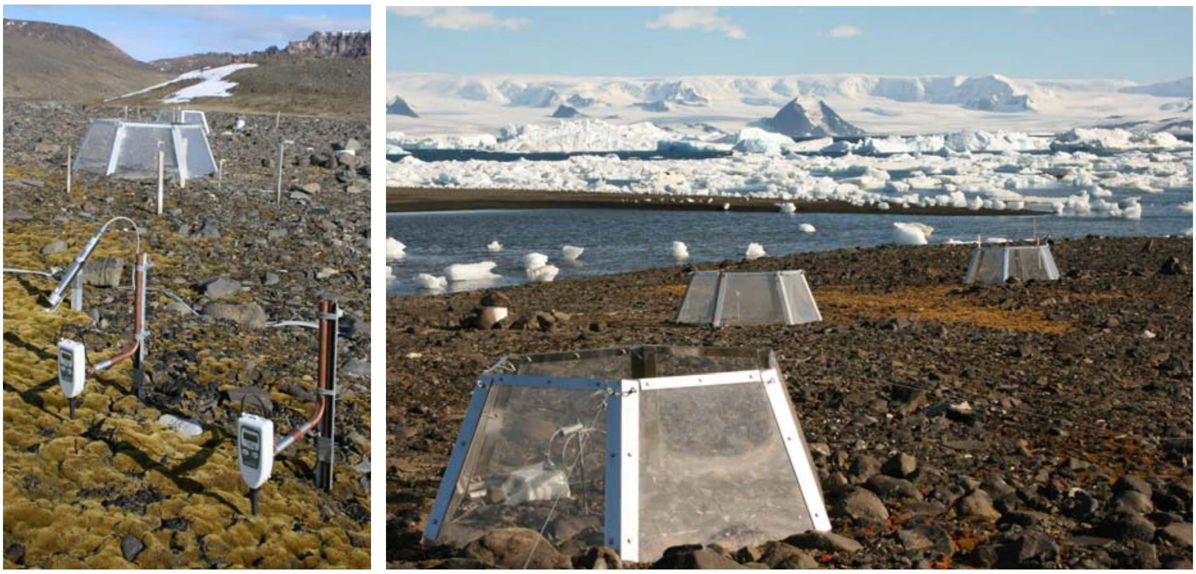

Fig. 2 Fluorometers installed at the outside control plots (left) and inside open top chambers located at the coastal area (I) close to the J.G.Mendel station, James Ross Island.

Results:

For 2007 and 2008 austral summer, analysis of OTC-induced microclimate is given in our forthcoming publication [8]. Here we report only major results for the OTCs located in the coastal area (referred as I) and those located at the top of table mountain (referred as II). In both locations, OTCs induced shift in air, surface and soil temperature when compared to outside control. In January-February 2007, there was apparent difference in OTC-induced air temperature shift measured at 30 cm height inside OTC between coastal location (I) and meseta (II). While mean daily shift reached 0.9 ^{o}C in coastal OTC, it was apparently higher (1.7 ^{o}C) at the mesa. The temperature shift in daily means was much higher for sunny calm days (1.6 and 2.5 ^{o}C for I and II, respectively) and apparently lower for overcast windy days (0.4 and 0.9 ^{o}C for I and II, respectively). Similarly, OTCs-induced shift in air temperature at 30 cm was recorded in January-February 2008. Mean daily shift were 1.1 ^{o}C and 1.4 ^{o}C in I and II locations, respectively. The temperature shift was again much higher for sunny calm days (1.6 and 2.0 ^{o}C for I and II, respectively) than overcast windy days (0.5 and 0.8 ^{o}C for I and II, respectively).

Discussion:

Analysis of temperature data shown, that air temperature inside the OTC incresed, more apparently in OTC located close to the seashore than those at mesa. As expected, the largest shift in temperature inside OTC against control plots was found during the days with sunny and calm days. Apart from direct solar radiation, wind speed was the main factor affecting the extent of temperature shift inside the OTC, since during windy days with Ws > 10 m s^{-1}, there was hardly any difference between inside and outside air temperature. On the other hand, during sunny days with no or limited wind, surface temperature inside/outside OTC was absolutely higher at mesa, since basaltic stones were warmed more than organic substrates in seashore location. Therefore, *Usnea antarctica* and *Umbilicaria decussata* had to cope with more pronounced range of temperature than mosses and lichens at seashore within a single day. In some periods (*e.g.* Feb. 1-Feb. 4, 2007), incresed air temperature caused snow melting insite OTC resulting in bare substrate, while there was still snow cover at control plots for a couple of days. Such situations had a cosequence to water regime inside OTC. We addressed a question, whether or not the temperature inside OTC is predictable from temperature data collected at a neighbouring weather station. From the analysis, it follows that in majority of cases, good relation exists between weather station (temperature at 2 m above surface) and OTC. In our data, we demonstrate the effect of OTC on surface and substrate warming, most apparent at the depth of 5 cm at a seashore OTC. This might have a consequence for promoted microbial activity of substrates. Recent studies indicate, that long-term studies exploiting manipulated warming are unavoidable to predict the likely changes in community structure, biomass production and stability of Antarctic vegetation under ongoing global climate change [10].

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Usnea species biomass evaluation on volcanic mesas at James Ross Island, Antarctica

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Introduction:

A long term ecological research program has been carried out in a coastal Antarctica on the James Ross Island in the vicinity of the Czech scientific station J.G. Mendel, NW part of the Weddell Sea (63° 48' 02" S, 57° 52' 57" W), in the austral season 2006-2007 and 2007-2008. Miocene alkaline volcanic rocks form monogenetic outcrops (volcanic fields and flat mesas at the height cca 300 – 650 m. a.s.l.), are main geomorphologic structures. Ecologically extreme but stable environment of volcanic mesas is the most suitable environment for development of rich communities composed of lichens. The communities are dominated by several species of fruticose lichens (*Usnea sphacelata Usnea antarctica, Usnea subantarctica*) with scarce foliose representatives (*Leptogium puberulum*). One part of an ecological research program is *Usnea* species biomass evaluation by different methods.

Material and Methods:

Lichen ground cover and diversity has been evaluated by several methods. On 38 experimental botanical square plots (50x50 cm) set up on three mesas (Berry Hill, Johnson Mesa, Lachman Crags) the following measurements and field works has been made: 1) measurement of density and length of lichen thallus, 2) digital photography with subsequent ground cover evaluation (Chips program for Image analysis), 3) ground micro topography measurements. Simultaneously, the lichen biomass has been harvested from 14 plots and ash free dry weight (AFDW), chlorophyll content and lichen thallus surface evaluated (scanning). The main aim of this work was to find out whether or not it is possible to measure (estimate) lichen biomass with a non-destructive method (measurements with botanical square plots and digital photography).

Results:

Complex and advanced statistical approach has been applied to data obtained by all above-specified measurement (expert in Ecology Data Analysis, Petr Šmilauer).

<u>LA (leaf area) determination:</u> The statistical models, in which digital photography and lichen thalli measurements are used, give the most accurate result in LA estimation. 82.3% of variability in the values is clarified and the correlation between fitted and real LA values is 0.944 and the accuracy of the LA estimation (50x50) cm is ±237 cm^2. Consequently, the "jackknife" method was used for more realistic LA estimation and the result is then ±423 cm^2. The significant deviations from the common model have square plots St_1_Berry, St_6_Berry and St_3_Bibby.

LA estimation determined only from the digital photography (Cover) of the experimental plot was not accurate; only 22.7% values variability was clarified by this approach. ±605.8, resp. 675.3 (jackknife) is the accuracy of the LA estimation.

Biomass determination: The same method was used for lichen biomass determination and again, both digital photography (Cover) and lichen thalli measurements had to be used for the calculation. In this case, 87.5% values variability was clarified and correlation between fitted and real values was 0.956.
Result, including only the digital photography (Cover) was similar to the LA determination, i.e. only 20,1% of values variability was clarified by this approach.

Chlorophyll *a* determination: It was not possible to estimate Chlorophyll *a* concentration from the measured values in this statistical test.

Chlorophyll *b* determination: Chlorophyll *b* concentrations were dependent on the experimental plot micro relief. 55.5% of the values variability was clarified and the correlation between fitted and real values was 0.768.

Glycolipid and pigment transformations of *Deschampsia antarctica* to UV-B irradiation

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Key-words: *Deschampsia*, UV-B irradiation, carotenoids, glycolipids.

Introduction:

The Antarctic geobotanical zone is a hostile environment for plant growth. *Deschampsia antarctica* Desv. *(Poaceae)* is the only native *Gramineae* found in the Antarctic. The species is naturally adapted to cold maritime Antarctic climate. Its range is restricted to the Antarctic Peninsula and its offshore islands [1, 2, 3]. Low temperatures, high light, stratospheric ozone depletion and enhanced solar UV-B irradiation induce the formation of reactive oxygen species (ROS) in high levels [4]. ROS negative effects in plants can be reduced by e.g. carotenoids (among other compounds) which react with free radicals forming carotenoids radical directly [5]. Carotenoids could be regenerated by interaction with tocopherols and ascorbate in the membrane lipid phase [6]. Thus, we study the carotenoids and glycolipids changes in the mechanisms of plant resistance to UV-B action, on the example of *D. antarctica* as a unique *in situ* indicator of global environment changes.

Materials and Methods:

D. antarctica plants were collected from some offshore Antarctical islands. Plants of *D. antarctica* with an intact root system were selected, with the help of colleagues of the 13th Antarctic Ukrainian expedition and supply to Ukraine in containers by air within 3-days.

The control plants were grown under the laboratory condition for 10 days under the lamps of daily illumination by air temperature 8-10 °C, 16-hour photoperiod and irradiated by UV-B for 20 hours in 5-times exposition (4h on light period). The UV-B lamp with absorption filter (TL 20Вт/12RS (Philips) 280-300 nm) was used for illumination of the plants. The biologically effective UV-B radiation (UV-BBE) was 6.17 kJ m^{-2} d^{-1}. Distance to the source of illumination was 10 cm.

The pigment content in leaves was determined with generally accepted method [7]. The separate carotenoid content was determined using TLC method [8] in our modification. Polar lipids were isolated according to L.Zill and E.Harmon [9] in modification of G.Yakovenko and A.Mihno [10]. Glycolipids were separated with the help of TLC and then monogalactosyldiacylglycerol (MGDG) and digalactosyldiacylglycerol (DGDG) were determined by densitometring of TLC plates against standards [11], sulfoquinovosyldiacylglycerol (SQDG) – according to E.Kean [12].

The replication of experiments was triple; authenticity of difference between the mean arithmetic values of indexes was set after the Student criterion. Differences were considered as reliable at the value of $p \leq 0, 05$.

Results:

The experimental UV-B irradiation caused significant changes in photosynthetic pigment content in leaves of *D. antarctica* plants. The UV-B treatment induced degradation of chlorophylls (Chls) *a* and *b*, but thereat Chl a/b ratio were unchanged (Fig. 1).

Analysis of carotenoid (Carots) composition revealed the absence of reliable changes in xanthophylls pool of UV-B treated *D. antarctica* plants. Following substances were identified: *β*-carotene, neoxanthin (Neo), violaxanthin (Vio) and fraction lutein (Lu) +

zeaxanthin (Zea). The content of Lu + Zea, Vio and Neo did not change under UV-B radiation. The UV-B induced only the degradation of β-carotene in the irradiated plants (Fig. 2).

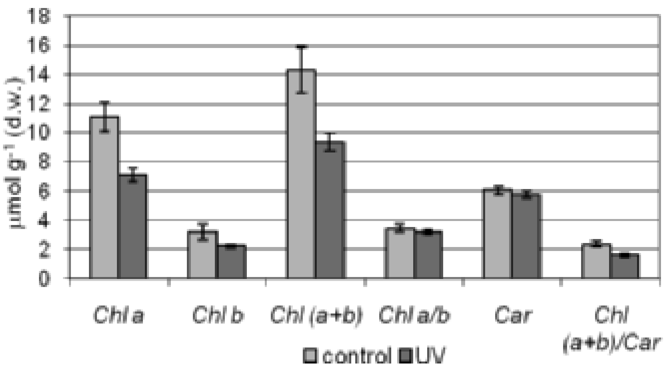

Fig. 1. Pigment content in *D. antarctica* leaves under UV-B irradiation

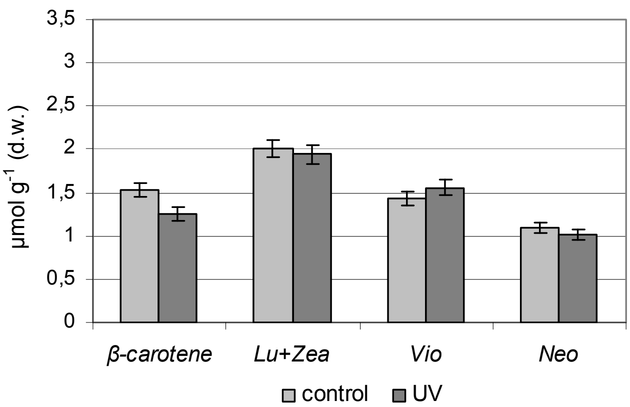

Fig. 2. Carotenoids content in *D. antarctica* leaves under UV-B irradiation

Glycolipid analysis demonstrated relatively stable SQDG content (but expressed as mol % even slight increase) in treated plants while there were significant MGDG content decrease and slight DGDG content decrease on 40 and 22% respectively (Fig. 3). These galactolipids transformations resulted in MGDG/DGDG ratio decrease.

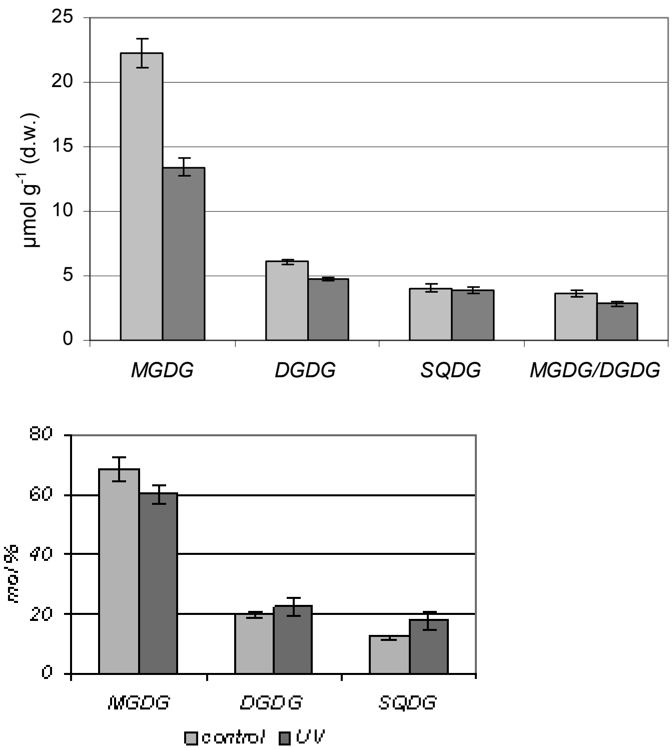

Fig. 3. Glycolipid composition of *D. antarctica* leaves under UV-B irradiation.

The ratio between MGDG and major xanthophylls could reflect transformation of lipid-pigment complexes during adaptation to stress conditions. Under UV-B irradiation MGDG/(Lu+Zea) ratio decreases to 38%, MGDG/Vio ratio to 45% (Fig. 4).

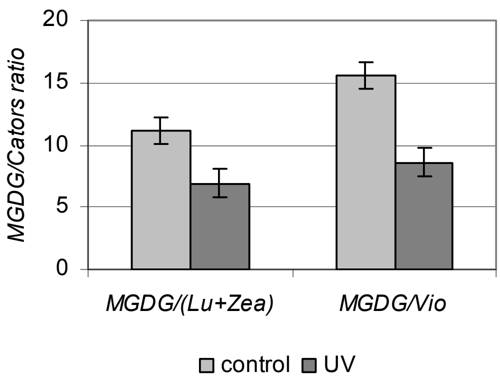

Fig. 4. The MGDG and carotenoids ratio of *D. antarctica* leaves under UV-B irradiation.

Discussion:

The *β*-carotene is present in antennae (LHCs) and reaction center (RC) of PSII and plays a major role in photoprotection by quenching the triplet state primary donor ($^3P_{680}$) or reducing the oxidized form (P_{680}^+) [13]. Molecules of *β*-carotene protect LHCs and RC against photooxidative damage through quenching of singlet oxygen ($^1O_2$) and/or triplet excited of chlorophyll ($^3Chl^*$) [14]. The mechanism of protection against $^1O_2$ leads to destruction of *β*-carotene molecules [15] and to some extent, is obstacle in the chain the oxidative reactions in plant cells [16].

The decrease of MGDG and DGDG content in UV-B treated plants can be considered result of degradation processes or related to lipoxygenase activation. MGDG/DGDG ratio decrease may indicate displacement of equilibrium between the bilayer and non-bilayer lipid structures of photosynthetic membranes. Enhancement of this ratio is the evidence of decreasing level of non-bilayer lipids and indication of less protection of chlorophyll-protein complexes to UV-B irradiation [17].

Our data match partly with results of T.Sakaki [18] study, who revealed a loss of pigments and lipids (mainly MGDG with some DGDG) as a consequence of oxidation stress caused by ozone exposure. However, the anionic lipid content was stable for the whole period of it in this case [18]. Concerning the meaning of the events observed in our study, we could suggest the stable SQDG amount being connected with energetic function of it. This compound seems to take part in adaptation reaction as cytochrome oxidase, CF1, F1, ATPase regulators, protectors and stabilising agents for D1/D2 dimers and LHCII [19, 20]. Early stages of photoinhibition induce degradation and cleavage of D1 protein of RC PS II [21]. It is possible that SQDG localised at the surface of the native D1/D2 heterodimer [22] hold monomers together as dimer [23] and stabilizes it while unfavourable environment changes.

Thus, the modifications of glycolipids and carotenoids pool were represented in change of lipid-pigment ratios, namely MGDG/Lu+Zea and DGDG/Vio. The decline of MGDG/DGDG ratio took place as result of the significant MGDG decrease while anionic lipid SQDG was stable in irradiated plants. These ratios characterise capacity of *D. antarctica* photosynthetic apparatus to adapt to UV-B irradiation at the level of lipid-pigment complexes.

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Comparative study of dehydration-response curves of photosynthesis in Antarctic lichen and *Nostoc commune*

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Key-words: chlorophyll fluorescence, effective quantum yield of photosynthesis in PS II, *Xanthoria* sp., *Nostoc commune*

Introduction:

Lichens are poikilohydric symbiotic organisms that tolerate desiccation. They survive in dry state by inactivation of their metabolic activity. After rehydration, they are capable to restore their physiological activity in terms of minutes. However, lichens are not able to control water content in their thalli. Thefore, they desiccate quite rapidly if exposed to drying conditions, such as e.g. low relative air humidity, high air/thallus temperature, windy weather. During desiccation, they keep their physiological activity even under gradual loss of water. It has been demonstrated that lichen maintain their photosynthetic activity unchaged at water potentials that are critical for higher plants [1,2]. Severe loss of water from a lichen thallus, however, leads to substantial loss of photosynthetic activity and, finally, full inhibition of photosynthetic processes [3] at critical water potential. Some authors [4] have pointed out that the water potentials ranging between -20 and -25 MPa are critical for the photosynthetic activity for majority of the lichen species. Experimental studies using natural atmospheric dehydration proved -20 MPa as critical value for a foliose lichen *Lobaria pulmonaria* [4]. Similarly, critical value of -18.8 MPa was found for *Lasallia pustulata* and *Umbilicaria hirsuta* exposed to osmotically-induced dehydration [5].

In Antarctic lichen species, responses of photosynthetical processes to dehydration and critical water content in lichen thalli were measured only sporadically, e.g. [6]. The aim of this study was to evaluate critical water potential for some Antarctic lichen species, so that interspecific differences could be found. Chlorophyll fluorescence technique was used to evaluate critical water potential because chlorophyll fluorescence parameters, effective quantum yield of photosynthetic processes in photosystem II (Φ_{PSII}) in particular, sensitively monitor dehydration-induced inhibition of photosynthesis [7].

Material and Methods:

Lichens (*Xanthoria* sp.) and *Nostoc communae* were collected in February 2009, during the Chech expedition to the James Ross Island in a close vicinity of J.G. Mendel station. After transport to the Czech Republic, the thalli were kept in dry state in dark. Before measurements, thalli were rehydrated by spraying and kept in fully hydrated state under the dim light conditions (up to 10 μmol m^{-2} s^{-1}) for 24 hours at room temperature. In order to determine the relation between the water potential and the quantum yield of PSII, an OPTI-SCI fluorometer (Optiscience, USA) was used. For water potential measurements, a WP4-T dewpoint water potential meter (Decagon Devices, US) was used. Simultaneous measurements of effective quantum yield of photosynthetic processes in photosystem II (Φ_{PSII}) and WP were made on gradually desiccating samples exposed to actinic light of PFD 60 μmol m^{-2} s^{-1} and at constant air temperature (18 ^{o}C) monitored by sensor/data logger (HOBO, Onset Computers, US). For each species, at least six replicates were measures.

Results:

Decrease in water potential (WP) from full saturation (WP=0 MPa) to -6 MPa did not lead to substantial loss in Φ_{PSII} in *Xantoria* sp. while gradual decrease of Φ_{PSII} was apparent in *N. commune* within the same WP interval. During following dehydration, at WP values below -6 MPa, both experimental species shown more or less constant rates of Φ_{PSII} decrease. Critical WPs, at which photosynthetic processes were severely limited by water loss and, finally, inhibited, were estimated -20 MPa for *Xanthoria* sp. and -30 MPa for *N. commune.*

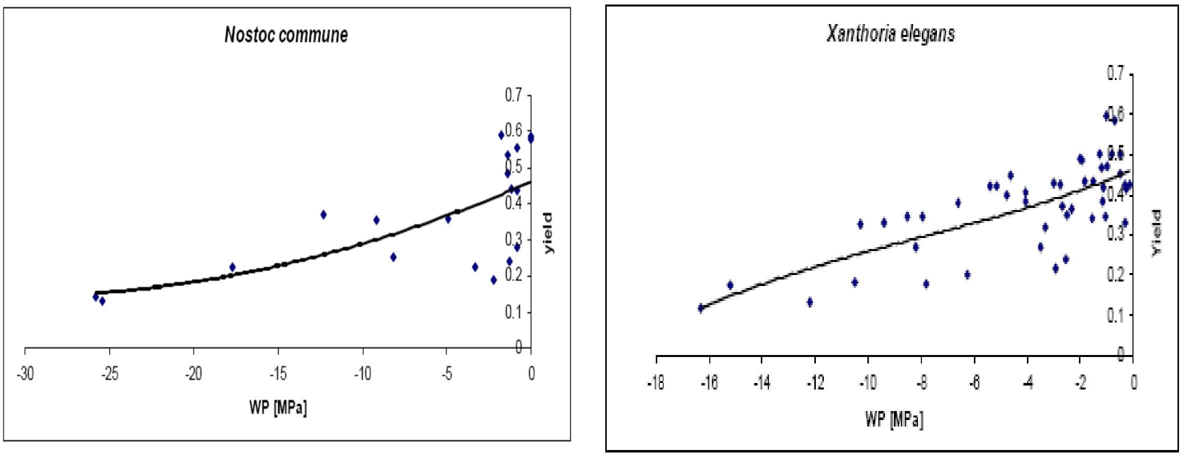

Fig. 1. The relationship between the quantum yield of Chl *a* fluorescence (Φ_{PSII}) and the water potential in nonlichenized *Nostoc commune*(left) and *Xanthoria* sp. (right).

Discussion:

The decrease in Φ_{PSII} with more pronounced desiccation is rather general phenomenon in lichens associated with dehydration-induced inhibition of primary photosynthetic processes. In PSII, it comprises mainly functional disconnection of LHCs from PSII core as stated e.g. by [8] and consequent decrease in light driven processes in the PSII center. The quantum yield of the lichen *Xanthoria* sp. has been previously measured and the results revealed biphasic relation between the Φ_{PSII} and the water potential [5]. The photosynthetic activity does not appear being limited during the early stages of lichen desiccation, but starts be inhibited when the values of water potential drop below -8 MPa. The findings of the relationship between the quantum yield (Φ_{PSII}) and the water potential in the lichen species examined herein by the above mentioned methodology are in a good agreement with the previous results [9].

Recent measurements of quantum yield performed at the non-lichenized *Nostoc commune* have revealed that the cynobacteria exhibited gradual loss of photosynthetic activity. The response of Φ_{PSII} to decerasing WP did not showed biphasic character, however, critical WP was found wery low comparably to *Umbilicaria antarctica* (M. Barták, unpublished data). Therefore, *N. commune* may retain its photosynthetic activity even at severe water loss which might be particularly advantageous in Antarctic habitats with low availability of liquid water.

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Isolation and characterization of novel cold-active hydrolyses from Antarctic streptomycetes

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Key-words: *Streptomyces sp*., Antarctic strain, cold-adapted enzymes, AZCL-Amylose, AZCL-Casein, protease, amylase

Introduction:

Enzymes have attracted attention of researchers all over the world because of wide range of physiological, analytical and industrial application. This is partially true for the enzymes extracted from microorganisms because of their broad biochemical diversity, feasibility of mass culture and ease of genetic manipulation. More than 3000 different enzymes have yet been identified and many of them found their way into biotechnological and industrial applications. The present enzyme toolbox is not sufficient to meet many industrial demands. Therefore, researchers are now trying to exploit extremophiles which are valuable source of novel enzymes [1].

Filamentous bacteria from *Streptomyces* genus exhibit remarkable capacity for the synthesis of secondary metabolites and use of numerous extracellular hydrolytic enzymes to degrade organic material in their natural habitat [1, 2]. For that reason, cold-active enzymes have important biotechnological applications in food industry, as well as biomass conversion, bioremediation, inasmuch as running processes at low temperatures reduces the risk of contamination by mesophiles and also saves energy [3, 4, 5, 6, 7].

The aim of this study was to isolate and characterised new polar streptomycetes capable of to biosynthesis of cold-adapted amylases and proteases. The reported enzymes may have wide spread applications for detergents, food industry and bioremediation processes at low temperature.

Material and methods:

Microorganisms: The selected 4Alga strain was isolated from Antarctic vegetation samples from Progress Lake 2 (69° 20' 00.0" S to 69° 29' 00.0" S/- 69.3333° to - 69.4833° 75° 55' 00.0" E to 76° 35' 00.0" E /75.9167° to 76.5833°, East Antarctica coast).

Identification of the strain was achieved by light microscopy, biochemical and cultural characterization. Biolog microtiter plate, a system with 96-wells and 95 different carbon sources was used to characterize the novel 4Alga strain by determining the ability to oxidize various carbon sources (http://www.biolog.com). The results were interpreted on a scale of +, positive reaction; -, negative reaction.

Enzyme characterization:

Enzymatic liquid culture was acquired during 10 days of submerged cultivation on liquid medium with following composition (g/l): soluble starch 20.0, corn steep liquor 10.0, $(NH_4)_2SO_4$ 6.0, $CaCO_3$ 8.0, NaCl 5.0 and soybean oil 0.2 ml, pH 7.0, [8], inoculated with 2% spore suspension and incubated at 20°C, at 230 rpm. After that the biomass was separated at 5000 rpm during 15 minutes. Using enzymatic liquid culture hydrolyses rate was tested at different temperature values (5-50°C).

As a basal medium, 2% agar medium was used at neutral pH, supplemented by adding 0.05% the insoluble chromogenic functionalized substrates based on AZCL. The commercial AZCL-Amylose and AZCL-Casein powers were transferred in 96% ethanol and added into the basal medium. To abide the particles dispersed, autoclaved medium was agitated gently being poured into plates. Wells into the medium were realised and than pipetted (25-35 µl liquid culture). In this sense, the diameter of hydrolyses zone, expressed in cm was appreciated.

Effect of temperature on hydrolase's enzyme production:

The temperature profiles were determined by assaying the enzyme activity at different temperatures 5-80°C (for alpha-amylase activity MIUG method was used, beta-amylase activity was realised using Merck method and for protease activity Anson method was used) [8].

Results:

The microscopic examination of the selected Antarctic isolate 4 Alga revealed that aerial mycelia were morphologically branched with compact spirals of sporophore. Outer surface of colonies were perfectly round initially, but later they developed aerial mycelium that may appear powdery and spore formation started after 4th day of incubation.

Biolog microtiter plate, 96-wells with 95 different carbon sources was used to characterize the novel 4Alga strain by determining the ability to oxidize various carbon sources. It was observed that 4 Alga strain was able to oxidize the arabinose, xylose, mannitol, L-phenylalanine. There are many reports on utilization of carbon sources, substrates that are certified also by [2]. Fructose, dextrin, glycogen, Tween 40, Tween 80, L-arabinose, L-aspartic acid, L-leucine, glycerol, citric acid, itaconic acid, D-mannitol were well utilized; sucrose, inositol, trehalose, formic acid, malonic acid, L-alanine, inosine were not utilized. Nevertheless, subsequent to microscopic examination, cultural and biochemical characteristics, it can be said that the selected isolates from Antarctic vegetation 4Alga belong to the *Streptomyces* genus. The temperature profiles were determined by assaying the at enzyme activity at different temperatures ranging from 5 to 80 °C.

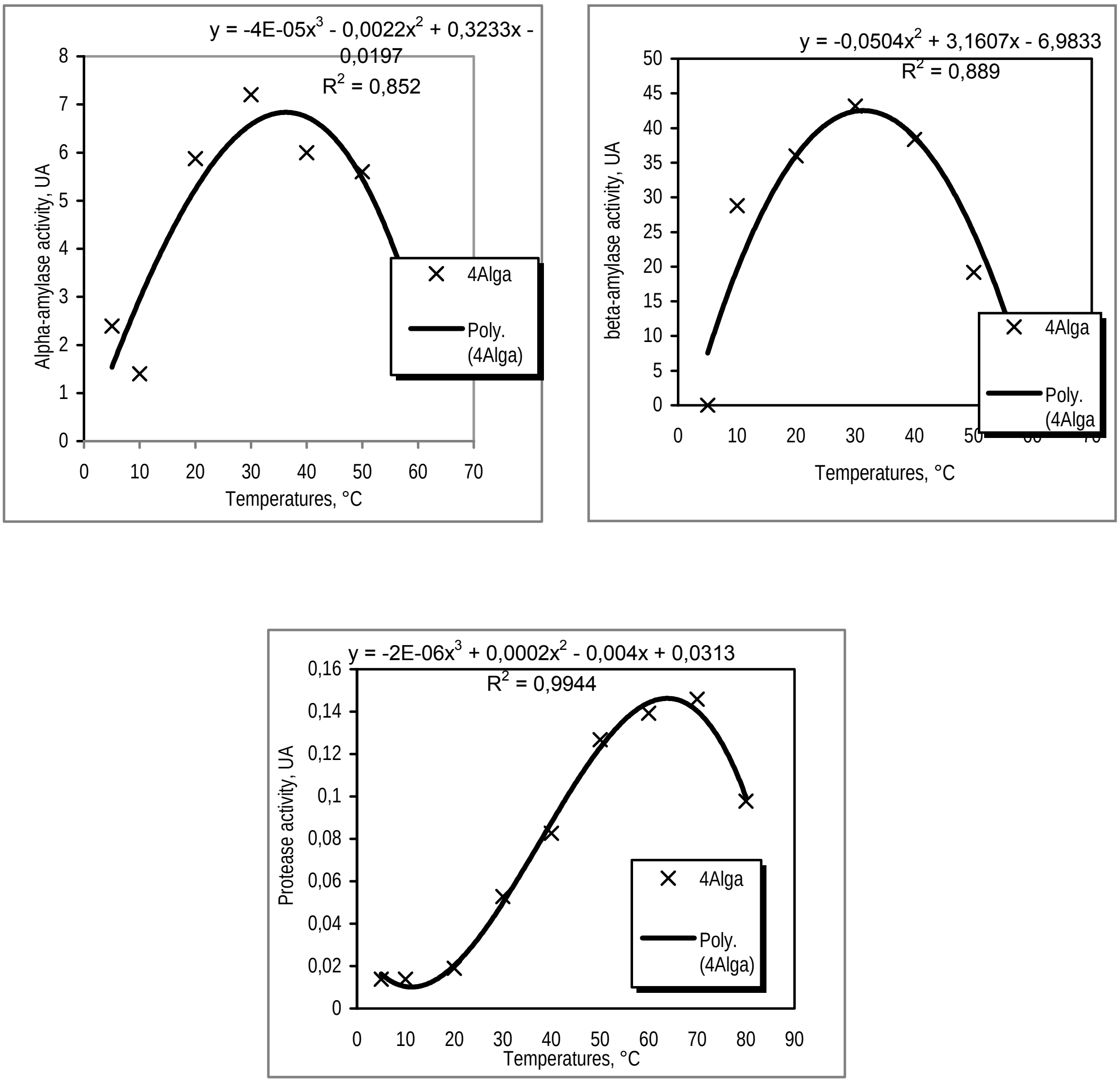

Fig. 1. Influence of temperature on hydrolase's activity produced by the 4 Alga streptomycetes

The results certify the ability of 4 Alga strain to produce amylase (alpha and beta amylase) and cold-adapted protease. The strain 4 Alga showed gradual increase in amylase and protease production but maximum amylase (alpha and beta) production was obtained at 30°C comparatively with the protease production where the optimum temperature was found at 70°C (Fig. 1).

Cold-adapted amylase and protease biosynthesis potential were evaluated regarding polar streptomycetes strains metabolizing insoluble chromogenic substrates based on azurine-crosslinked with amylose or casein (AZCL-Amylose, AZCL-Casein), detectable by the blue circles around the colonies. If the bacteria produce amylases or proteases, the enzymes hydrolyse the large substrate insoluble molecules, which have been dyed with AZCL. The small hydrolyzed compounds are still dyed blue and diffuse in the plate developing blue circles zones around the colonies.

The insoluble chromogenic substrate (AZCL-Amylose and AZCL-Casein) was degraded by 4Alga strain. It can be seen the blue halo surrounding the colonies. This phenomenon indicates that *Streptomyces* 4Alga had produced hydrolases at all studied temperatures. The diameter of hydrolyses zone was measured and express in cm.

Strain	AZCL-Amylose																	
	24 h						48 h						72 h					
	5	15	20	28	37	50	5	15	20	28	37	50	5	15	20	28	37	50
4Alga	0.7	0.8	0.8	0.9	0.8	0.5	0.8	1	1	1	0.8	0.6	0.9	1.1	1	1	0.8	0.6
Strain	96 h						120 h						144 h					
	5	15	20	28	37	50	5	15	20	28	37	50	5	15	20	28	37	50
4Alga	1.1	1.2	1.1	1	0.8	0.6	1.1	1.2	1.1	1	0.8	0.6	1.1	1.2	1.1	1	0.8	0.6

Table 1. Hydrolyses rate on AZCL-Amylose at 4 Alga strain at different temperatures

The data showed that *Streptomyces* 4 Alga strain was able to produce amylase active at low temperature (5, 15, 20°C), the maximum capacity of insoluble chromogenic substrate bioconversion being detected after 96 h of enzyme action (Table 1).

Strain	AZCL-Casein																	
	24 h						48 h						72 h					
	5	15	20	28	37	50	5	15	20	28	37	50	5	15	20	28	37	50
4Alga	0.4	0.5	0.6	0.7	0.9	1.1	0.6	0.8	0.8	1	1.5	1.5	0.7	0.9	1	1.3	1.7	1.9
Strain	96 h						120 h						144 h					
	5	15	20	28	37	50	5	15	20	28	37	50	5	15	20	28	37	50
4Alga	0.9	1	1.1	1.4	1.9	2.1	1	1.1	1.2	1.6	2.1	2.3	1.1	1.1	1.2	1.9	2.2	2.4

Table 2. Hydrolyses rate on AZCL-Casein at 4 Alga strain at different temperatures

Although 4 Alga strain showed the availability to grow at lower temperature and produce cold-adapted amylase. In protease case, the enzymes showed superior activity at 50°C, the biggest hydrolyze rate being developed after 144 h of incubation (Table 2).

The results certify the streptomycetes ability to produce amylases and cold adapted proteases. Studied polar streptomycetes having polymers-degrading activities can be successfully used in bioremediation process, dairy industry, bakery and detergents making, at low temperatures.

Discussion:

This work describes the capacity of new isolated *Streptomyces* 4 Alga strain to produce cold-active hydrolase’s using as a basal medium 2% agar medium adjusted with insoluble chromogenic functionalized substrates based on AZCL (azurine-crosslinked with amylose or casein). This technique provides a specific, reliable and rapid simultaneous detection of high active hydrolase’s producing strains.

The new isolated *Streptomyces* 4 Alga strain is highly able to biosynthesize amylases and proteases cold-adapted at low temperatures (from 5 to 20ºC). The studied enzymes, alpha-amylase and cold-adapted beta-amylase are active at 30ºC, meanwhile the proteases are active at 70ºC. These enzymes may have tremendous applications in detergents, bioremediation process, dairy industry, bakery, at low temperatures.

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Cryoproective effects of ribitol on lichen photosynthetic processes

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Introduction:

Lichens are extremophilic organisms capable to cope with majority of physical stressors. Among them, low and/or freezing temperature represents a factor to which physiological processes must be adjusted. It is well established that photosynthesis is detectable in Antarctic lichens even under – 20 ^{o}C (Kappen *et al.* 1996). However, low temperature limitation of photosynthesis exists at freezing temperatures because majority of lichen species have their temperature optimum for photosynthesis far above 0 °C (*e.g.* Hájek *et al.* 2001). In the field, majority of liches are in dehydrated, and thus physiologically inactive state at freezing temperature. However, when freezing temperature co-acts with fully hydrated lichen thalli, then the role of anti-freezing bodies is essential. In lichens, polyols (sugar alcohols) have several physiological roles. One of them is that sugar alcohols are effective anti-freezing substances (ribitol, arabitol, mannitol). Roser *et al.* (1992a) reported varying ribitol and mannitol concentrations up to 10 and 19 %, respectively, of total soluble sugars in thalli of Antarctic lichen species collected in different sites. Amount of total polyols and sugars in lichen thalli vary in different species and might reach up to 36 mg g^{-1}(d.m.) as reported by Roser *et al.* (1992b) for *Umbilicaria decussata* grown in maritime Antarctica. In this study, we bring an overview of ribitol effects on photosynthetic processes in lichens exposed to low and freezing temperature.

Material and Methods:

In several experiments, we tested a range of ribitol concentration added to lichen thalli exposed to low and subzero temperature. Using Chl fluorescence technique (a Fluor-Cam HFC-010, Photon Systems Instruments, Czech Republic), we detected a change induced by the addition of ribitol to cultivation Petri dishes. The aim of the study was to address the inter-specific differences between three foliose lichens: *Lasallia. pustulata* (L.) Mérat., *Umbilicaria. hirsuta* (Sw. ex Westr.) Hoffm., and *Xanthoparmelia somloensis* (Gyelnik) Hale. We hypothesized that addition of ribitol would maintain primary photochemical processes of photosynthesis active at subzero temperatures in lichen thalli, while the untreated thalli would exhibit temperature-dependent inhibition.

Thalli segments were cultivated in ribitol concentration of 8, 16, 26 mM for 72 h and 32, 50 mM for 168 h at temperatures +5, 0, and -5 °C. The chlorophyll fluorescence parameters (potential yield of photochemical reactions in PS II (variable to maximum fluorescence ratio, F_V/F_M), effective quantum yield of photochemical reactions in PS II (Φ_{PSII}), and non-photochemical quenching (NPQ) were monitored in 24-h intervals using a chlorophyll fluorescence imaging system.

Results and Discussion:

Addition of ribitol showed temperature-dependent response. In – 5 °C, externally added ribitol led to a positive change in chlorophyll fluorescence parameters. In majority of species, concentration-dependent increase in F_V/F_M, decrease in non-photochemical quenching (NPQ) but no change in quantum yield of photosystem II (Φ_{PSII}) values was found at –5 °C. At higher temperature (0, +5 °C), no effect of ribitol addition on the photosynthetic parameters was apparent. Such findings indicate cryoprotective role of ribitol. It is discussed in more details in Hájek *et al*. 2009a and Hájek *et al.* 2009b (*in press*). Surprisingly, 50 mM ribitol concentration treatment led to a decrease in F_V/F_M and Φ_{PSII} and to an increase in NPQ values at -5 °C, while no change was observed at 0 °C and +5 °C. This indicates inhibitory effects of ribitol, if its concentration is too high in a lichen thallus. Under natural conditions, however, ribitol is metabolized in lichen thallus quite fast and thus such high concentration (equivalent to 50 mM) is not realistic in the field. Moreover, ribitol is synthesized in algal partner and exported to fungus, where is transformed into mannitol by a pentose phosphatase pathway.

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Reflectometric study of structural and functional characteristics of Antarctic lichens

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Introduction:

Remote sensing and analysis of diffuse spectral reflectance, which has already been used frequently for monitoring of canopy structure of herb vegetation and for detection of physiological status of crop plant stands or forests [1], could be very helpful also in studies of polar vegetation dominated by lichens. But the thalli of lichens are structures substantially different from plant leaves in many respects. Their upper surface is not covered by a transparent cuticula and epidermis, but by a *cortex,* formed by a dense aggregation of fungal hyphae, often dark pigmented and with many air-filled spaces when not in fully water saturated state. It was not clear, to what extent the particular properties of the upper cortex might interfere with assessment of pigments and water content of deeper internal structures. An extensive field study of spectral reflectance indices using wide range of Antarctic lichen species in different hydratation state was realized with the aim to assess applicability of this non-destructive approach for monitoring of structural and functional status of lichen stands.

Material and Methods:

Spectral reflectance measurements were done with a reflectometric set UNISPEC (*PP Systems, USA*) equipped with a detector *MMS1/NIR enhanced (300-1100 nm)*, with an internal light source (halogen lamp), and bifurcated foreoptics with a special clip for stabilisation of the foreoptics orientation at a 60° angle to the sample plane. To calculate reflectance, spectral radiance scans of the samples were divided by radiance scans of a 99% reflective standard. The following reflectance indices were derived from spectral reflectance curves (**R** denotes reflectance and the subscripts refer to specific spectral wavelength):

- normalized difference vegetation index, NDVI = $(R_{900}-R_{680}) / (R_{900}+R_{680})$.
- structural independent pigment index, SIPI = $(R_{800}-R_{445}) / (R_{800}-R_{680})$.
- water index, WI = R_{900}/R_{970}

The measurements were done in samples taken from about 20 most abundant lichen species of different morphological types (fruticose, foliose, crustose) growing in the vicinity of research bases Vernadsky (Galindez Island) and J.G.Mendel (James Ross Island). Structural characteristics of the measured samples (water saturation deficit, chlorophyll content) were done in laboratory as described in [2].

Results and Discussion:

Detection of structural and physiological status of all tested lichen species using spectral reflectance analysis was sufficiently reliable within broad range of intrathalline water deficiency and even at high content of photoprotective metabolites in the upper cortex. The NDVI index was sufficiently suitable for assessment of vitality and chlorophyll content of the lichen photobionts. Reflectance signal in the near infrared region (at 970 nm) has been found as the most reliable water status indicator for the tested lichen species. Sensitivity of the spectral detection of water status was rather small at high hydration level (water saturation deficit < 20 %, or water potential > -1 MPa), but this is not much limiting its value and potential use, because physiological processes in lichens are usually inhibited at much lower values of water potential than in leaves of vascular plants. No significant changes in SIPI were found in the tested lichens during desiccation, which may be considered as an indirect indicator of chlorophyll stability in the symbiotic photobionts.

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