

Presentation of two Projects dealing with Microalgae:

**1. co-Operation SKD ‘COIN’
(FFG/Applied Research)**

**2. Metabolomics and Biodiversity of
Snow and Ice Phototrophs
(FWF/Basic Research)**

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**UNIVERSITY
OF APPLIED SCIENCES
UPPER AUSTRIA**



Research Team Biosciences

Our two fundaments are:

1. “We *make* them doing!”

► Molecular Biology: modifying fungi or cyanobacteria to produce valuable biomolecules

2. “We gonna *find* those which can do it!”

► (Exotic) microorganisms producing interesting metabolites. Like secondary compounds which they need for survival.

Applied use of Algae

- Food, nutritional supplements
- Pharmaceutic or cosmetic ingredients
- Production of valuable metabolites
 - antioxidants
 - pigments
 - fats (mainly polyunsaturated fatty acids)
 - vitamins etc.
- Biomass usage (e.g. biogas production???)
- Future research: biodiesel made from algae???

Where we can find algae?

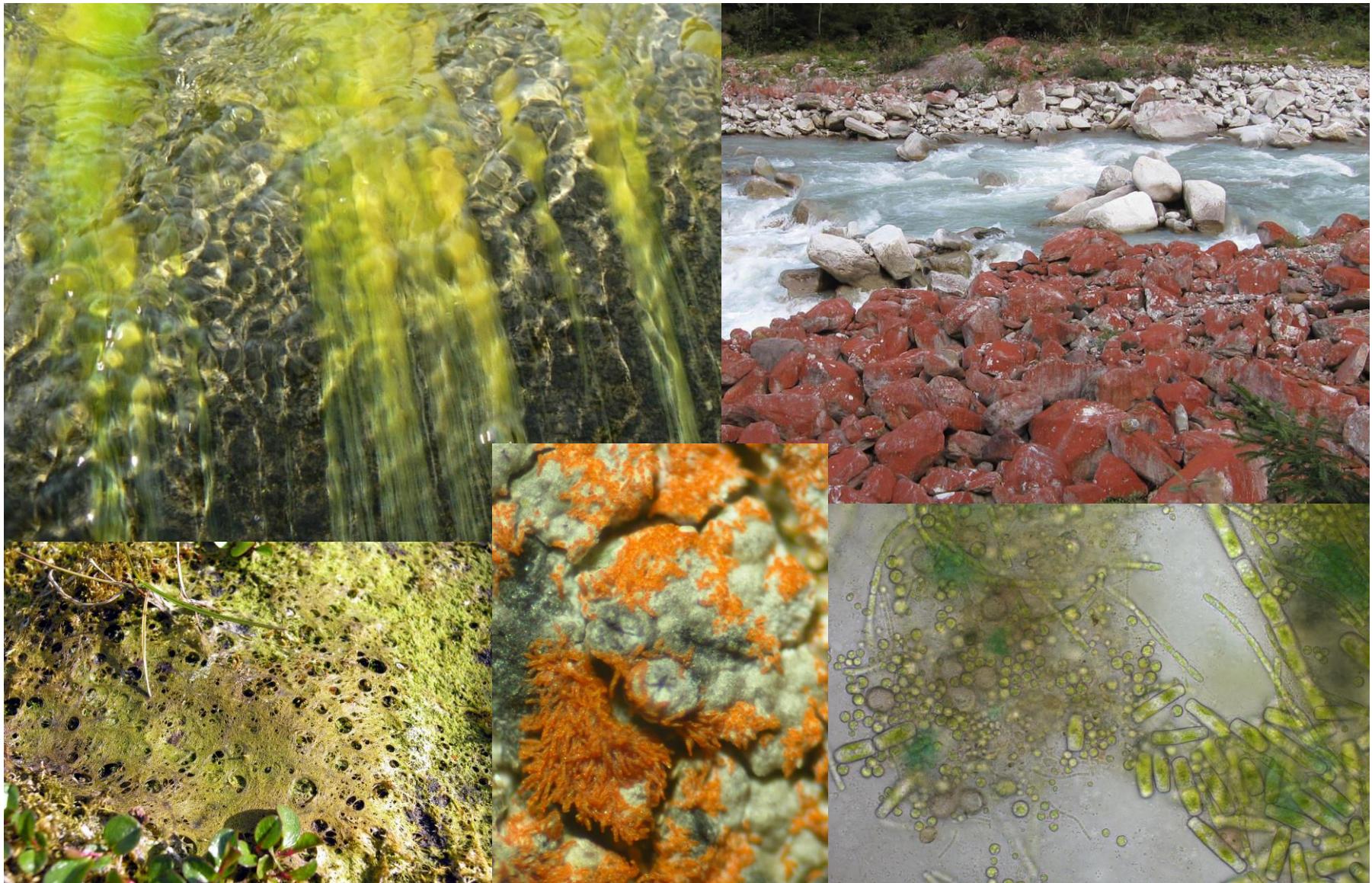


- **Habitats**
 - Marine (plankton, seaweed/macroalgae)
 - Freshwater (brooks, lakes, ponds, ...)
 - Specialized species also at rocks, tree bark or even at glaciers
 - Lichens: Symbiosis with fungi



Spitsbergen, Arctic

Algae: photoautotrophic eukaryotes



1. co-Operation SKD ‘COIN’ (FFG / Applied Research)



Head of Project in Wels: DI (FH) Klaus Krennhuber

- Strain collection of unexplored microalgae isolated from high alpine soils in the 1960/70ies (mainly Tyrolean Alps)
- Gaining competence in culturing strains, analyzing algal compounds and bioactivity assays
- Duration: 2016 - 2019
- Project partners: FH Wels, MCI Innsbruck, ADSI Innsbruck



co-Operation SKD 'COIN'

Illuminated & aerated
1 litre column bioreactors

Strains are evaluated in
these mini-reactors
(growth, yield, stress
experiments)

Stress in means of e.g. nutrients
(nitrogen-limitation), temperature
or salinity



Enzymaktivität

5 Kulturen wurden auf potentielle Enzymaktivität vermesssen. *Chlorella vulgaris*, *Chlamydomonas culleus*, *Spongiochloris* sp., *Diplosphaera chordatii*, *Chloroidium* sp.

Lipaseaktivität konnte bei *Chlamydomonas culleus*, *Spongiochloris* sp. identifiziert werden (unverifiziert)

Proteaseaktivität konnte bei *Diplosphaera chordatii*, *Chloroidium* sp. quantifiziert werden (verifiziert)

Produktanalytik

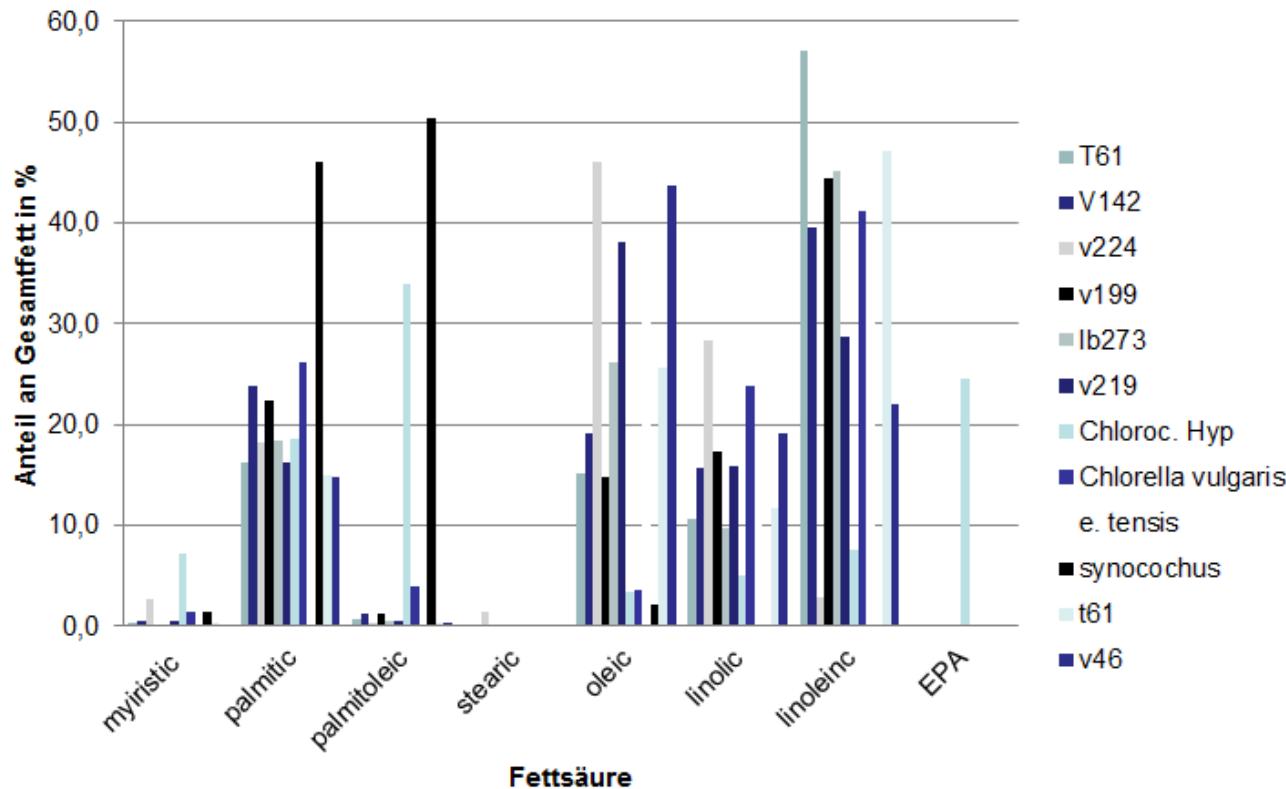
- Proteaseassay - Azo Casein-Test
- Azo Farbstoff, gekoppelt an Casein wird durch Protease in lösliche Länge gebracht; Färbung der Probe entspricht Enzym - Aktivität

Tabelle 1: Proteaseaktivität unterschiedlicher Algen bei Azo Casein Test in mU g⁻¹ Trockenmasse

<u>Algenstamm</u>	<u>Aktivität</u>
V199	337
V142	193
T61	766
V219	361
C. vulgaris	388

Fettsäurezusammensetzung

Verteilung der Fettsäuren in Algenproben



2. Metabolomics and Biodiversity of Snow and Ice Phototrophs (FWF/Basic Research)



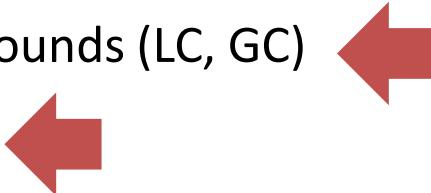
Head of project: Dr. Daniel Remias

Coastal Antarctica

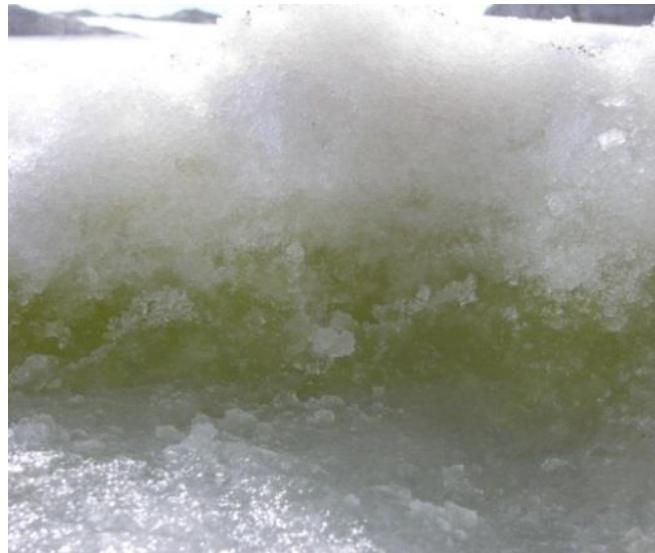
Preliminary publications about physiology and ecology of snow and ice algae

- Procházková, L., Remias, D., Řezanka T., Nedbalová, L. (2017) *Chloromonas nivalis* subsp. *tatrae*, subsp. nov. (Chlamydomonadales, Chlorophyta): re-examination of a snow alga from the High Tatra Mountains (Slovakia). *Fottea, accepted*
- Remias, D., Pichrtová, M., Pangratz, M., Lütz, C., Holzinger, A. (2016) Ecophysiology, secondary pigments and ultrastructure of *Chlainomonas* sp. (Chlorophyta) from the European Alps compared to *Chlamydomonas nivalis* forming red snow. *FEMS Microbiology Ecology, early online*, DOI: [10.1093/femsec/fiw030](https://doi.org/10.1093/femsec/fiw030)
- Remias, D., Jost, S., Boenigk, J., Wastian, J., Lütz, C. (2013) *Hydrurus*-related golden algae (Chrysophyceae) cause yellow snow in polar summer snowfields. *Phycological Research* 61: 277-285. DOI: 10.1111/pre.12025.
- Aigner, S., Remias, D., Karsten, U., Holzinger, A. (2013) Unusual phenolic compounds contribute to ecophysiological performance in the purple-colored green alga *Zygogonium ericetorum* (Zygnematophyceae, Streptophyta) from a high alpine habitat. *Journal of Phycology* 49: 648-660. DOI: 10.1111/jpy.12075.
- Remias, D., Wastian, H., Lütz, C., Leya, T. (2013) Insights into the biology and phylogeny of *Chloromonas polyptera* (Chlorophyta), an alga causing orange snow in Maritime Antarctica. *Antarctic Science* 25: 648-656. DOI: [10.1017/S0954102013000060](https://doi.org/10.1017/S0954102013000060)
- Remias, D. (2012) Cell structure and physiology of alpine snow and ice algae. In: Lütz, C. (Editor) Plants in alpine regions. Cell physiology of adaption and survival strategies. Springer Wien, 202 p., pp. 175-186. ISBN 978-3-7091-0135-3
- Remias, D., Schwaiger, S., Aigner, S., Leya, T., Stuppner, H., Lütz, C. (2012) Characterization of an UV- and VIS-absorbing, purpurogallin-derived secondary pigment new to algae and highly abundant in *Mesotaenium berggrenii* (Zygnematophyceae, Chlorophyta), an extremophyte living on glaciers. *FEMS Microbiology Ecology* 79: 638-648.
- Remias, D., Holzinger, A., Aigner, S., Lütz, C. (2012) Ecophysiology and ultrastructure of *Ancylonema nordenskiöldii* (Zygnematales, Streptophyta), causing brown ice on glaciers in Svalbard (high arctic). *Polar Biology* 35: 899-908.
- Sattler, B., Post, B., Remias, D., Lütz, C., Lettner, H., Psenner, R. (2012) Cold Alpine Regions. In: Bell, E.M.: Life at extremes. Environments, Organisms and Strategies for Survival. Wallingford CAB International Publishing, pp. 138-154. ISBN 978-1-84593-814-7
- Remias, D., Albert, A., Lütz, C. (2010) Effects of realistically simulated, elevated UV irradiation on photosynthesis and pigment composition of the alpine snow alga *Chlamydomonas nivalis* and the arctic soil alga *Tetracystis* sp. (Chlorophyceae). *Photosynthetica* 48: 269-277.
- Remias, D., Karsten, U., Lütz, C., Leya, T. (2010) Physiological and morphological processes in the Alpine snow alga *Chloromonas nivalis* (Chlorophyceae) during cyst formation. *Protoplasma* 243: 73-86.
- Leya, T., Rahn, A., Lütz, C., Remias, D. (2009) Response of arctic snow and permafrost algae to high light and nitrogen stress by changes in pigment composition and applied aspects for biotechnology. *FEMS Microbiology Ecology* 67: 432-443.
- Remias, D., Lütz, C. (2007) Characterisation of esterified secondary carotenoids and of their isomers in green algae: a HPLC approach. *Algological Studies* 124: 85-94.
- Remias, D., Lütz-Meindl, U., Lütz, C. (2005) Photosynthesis, pigments and ultrastructure of the alpine snow alga *Chlamydomonas nivalis*. *European Journal of Phycology* 40: 259-268

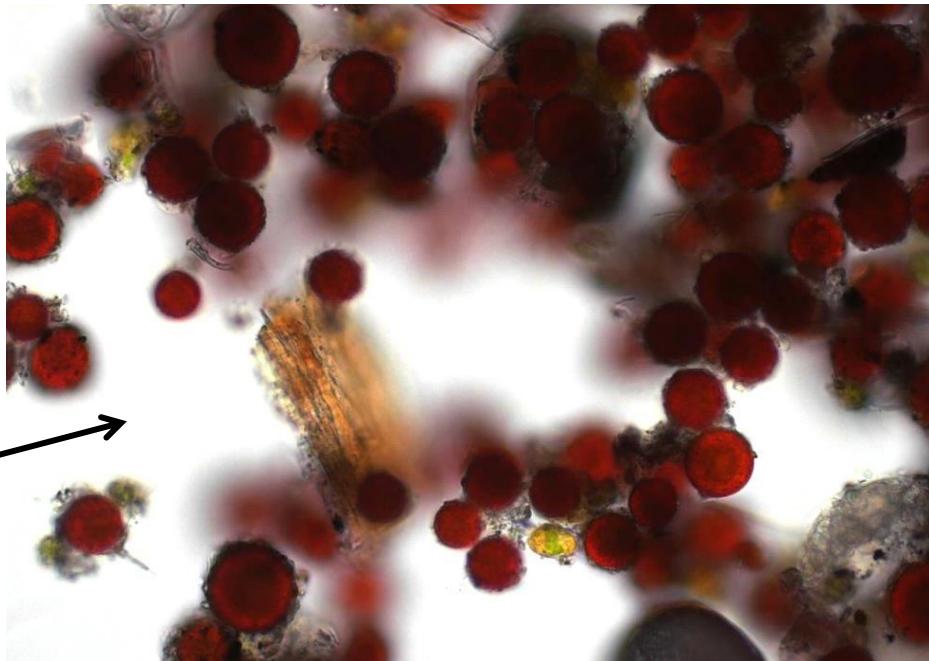
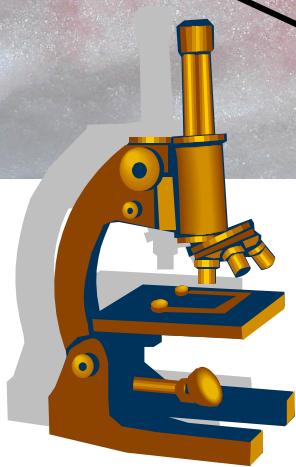
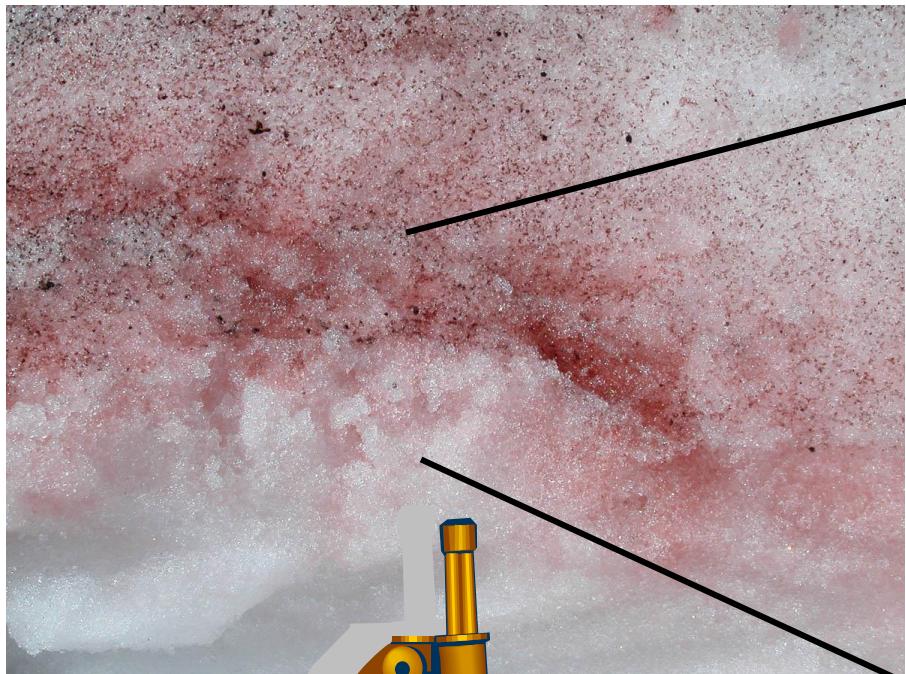
Metabolomics and Biodiversity of Snow and Ice Phototrophs

- Project start: April 2016
 - Duration: 3.5 years
 - Project sum: 350.000 €
 - People at Wels: post-doc, technician, master students
 - Project partners: Helmholtz Zentrum Potsdam (DE), Universities of Innsbruck (AT) and Prague (CZ)
 - Project aims:
 - Biodiversity evaluation (molecular Polar-Alpine comparison)
 - Metabolomics of field samples (GC-MS)
 - Screening for interesting (secondary) compounds (LC, GC)
 - Establishing new strains for biotechnology
- 

What are your associations looking at these snow colors?

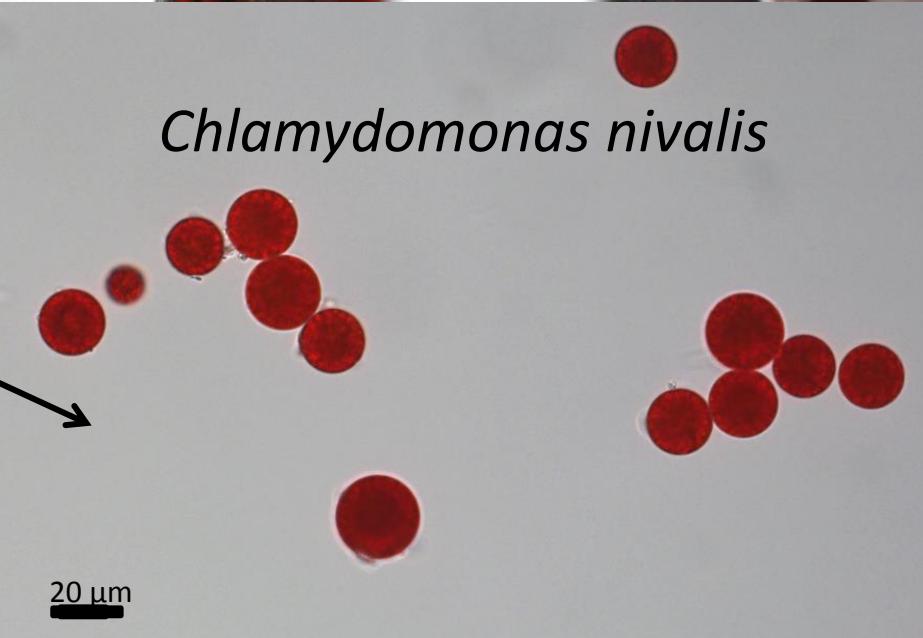


Tinted snow is caused by
microalgae



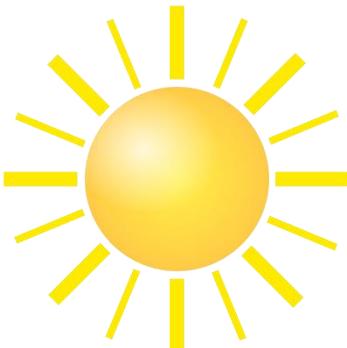
Chlamydomonas nivalis

20 μm



Snow algae have to cope with a lot of stresses

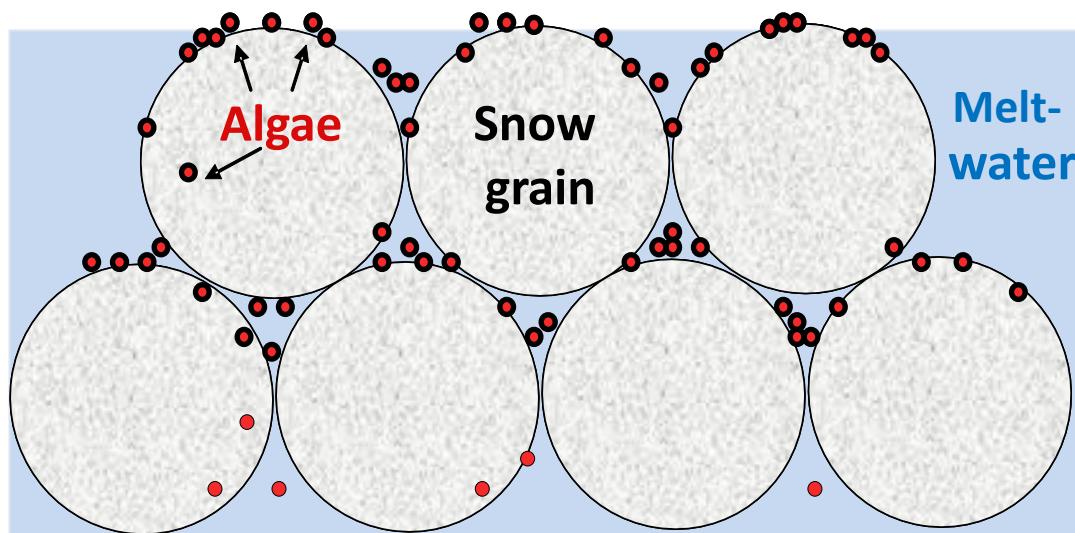
Excessive irradiation
(visible & UV)



Freeze-thaw cycles
(day / night)



Short season
(weeks/year)



Limited nutrients
(e.g. N, P)

Why to work with snow algae?

Explore biodiversity of
polar and high alpine cold
ecosystems

Understand fundamental
strategies of stress
response

Global warming supports
algae reducing the albedo of
Greenland's ice cap:
faster melt → rising sea-level

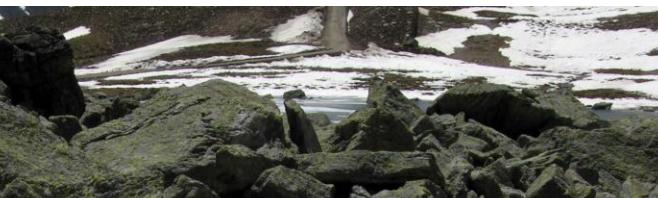
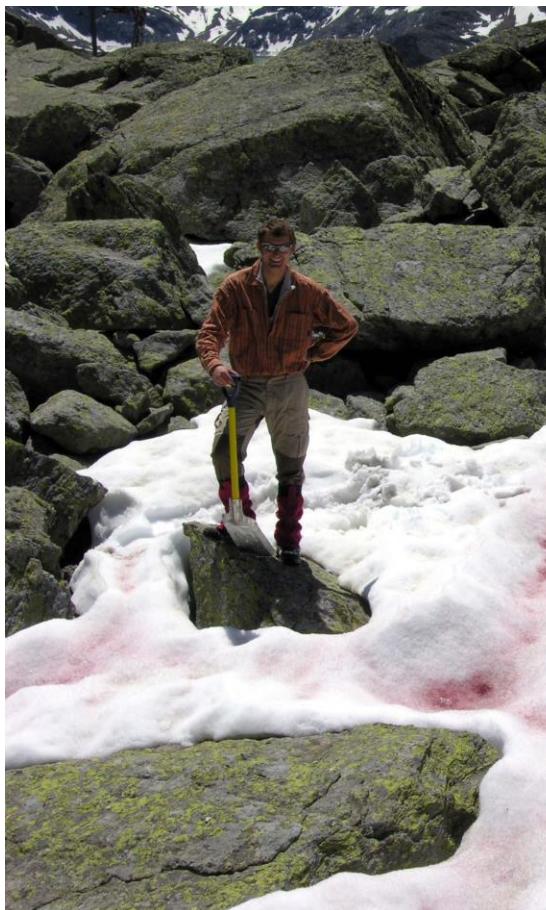


Special metabolites could
be beneficial for
biotechnology/applied
fields



UV screening compounds,
anti-freezing metabolites
(compatible solutes)
unsaturated fatty acids
anti-stress proteins

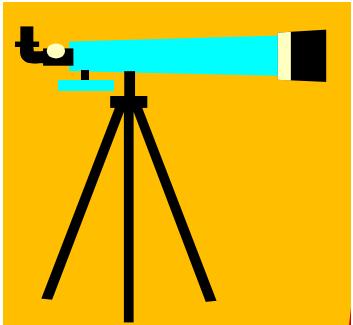
No strains available to work with? Harvest of field material!



Special equipment: low temperature climate chamber (Perceval LT-36VL)

- Stable down to -10°C (\pm 0.5) while illuminated!
- Needed for cultivation of psychrophilic strains





Main methodical approaches

Arctic-alpine comparison of cryoflora

- High Throughput Sequencing (18S, *rbcL*, ITS2 genes)

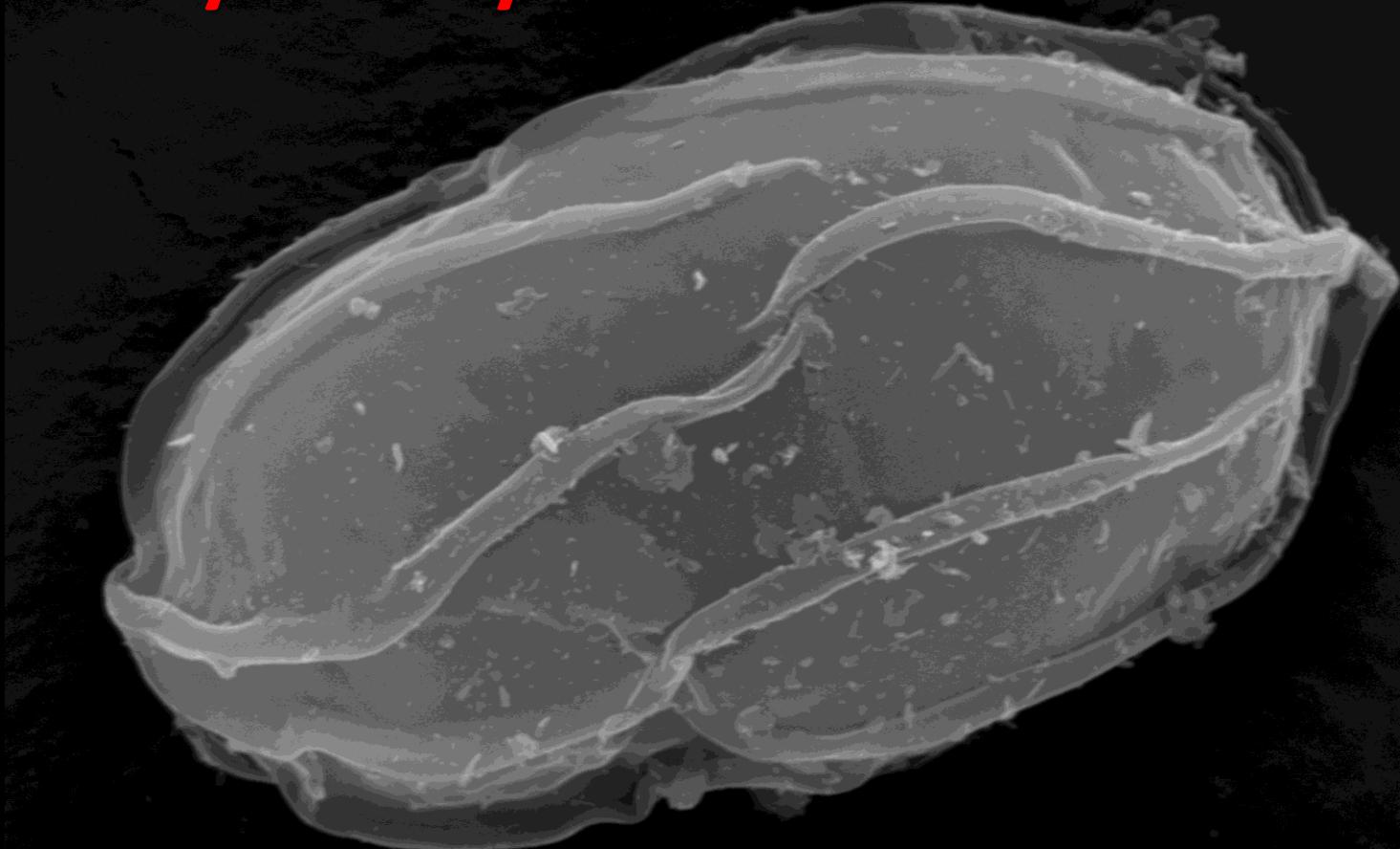
Evaluate presence of all eukaryotes like algae and fungi, but also bacteria and archaea. Look for geographical patterns (Helmholtz Potsdam)

- Metabolomics (GC/MS)

Gives ‘snapshot’ of the physiology of the cell. Cellular processes cause biochemical fingerprints (Innsbruck University)



Thank you for your attention!



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