# Rapport fra «AeN workshop in metabarcoding analyses»

## Innhentede svar pr. 13. januar 2020 15:00

- Leverte påmeldinger: **9**
- Påbegynte påmeldinger: **0**
- Antall invitasjoner sendt: **0**

**Your name (first name and surname)? ***

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**Describe briefly your research project. ***

- My research is focused on describing taxonomic and functional biodiversity from inland waters (i.e. lakes, ponds) in arctic and subarctic localities.
- We will conduct various microcosm experiments (exclusion of grazers of different size classes and addition of different DOC sources) during the four seasonal AeN cruises, where we are interested in investigating the changes in the food web as well as the changes on the prokaryotes and protists. Our group has established a simplified “minimum” microbial food web model (Thingstad et al., 2007). Here bacteria, autotrophic flagellates and diatoms (i.e. the osmotrophs) compete for mineral nutrients (N & P). The bacteria and diatoms are in addition controlled by supply of DOC and Si respectively, while the autotrophic flagellates and the diatoms also depend on light. The predators, heterotrophic flagellates, ciliates and mesozooplankton (i.e. the phagotrophs) each exert a top-down control on smaller forms and constitute a grazing food chain. Manipulating top predator level and nutrient conditions should give insight on how the system is controlled, how the groups interact (top-down, bottom-up, cascading effects) and how the nutrients flow through the system. Besides abundance and activity measurements we are interested in potential community composition changes due to the different treatments, which we intend to analyse using molecular methods including Illumina amplicon data.
- I am using a common garden setup to analyze the effect of habitat-specific fungal communities on the establishment and growth of mountain birch (Betula pubescens). I am cultivating birch seedlings in soil collected from various mountain plots above the climatic tree limit along a climate gradient in Norway and will use metabarcoding approaches to analyze the fungal community composition in the soil before/after 3 months of B. pubescens grown in addition to a metabarcoding analyses of the birch roots to find which potential mycorrhizal symbionts estalished during the early phase of growth. The goal is to see if the fungal communities above the tree-line will inhibit the establishment of birch once the rising temperatures in the mountains increase the altitude of the climatic limit for trees.
- I am mapping rare species of fungi using metabarcoding of soil samples in selected nature types. This data will be compared to fruitbody survey data in the same forests, thus testing the validity of metabardcoding for detection of rare species in soil.
- protist diversity, seasonality and activity using genetic methods (metabarcoding, metatranscriptomics, qPCR)
- Going to use 18S metabarcoding as a tool for diet analysis in small invertebrates sampled during the seasonal cruises. Planning to use R and the DADA2 pipeline. Small copepods (e.g. Oithona, Microsetella, Microcalanus) are likely to be the first target species, but benthic/sympagic nematodes (and thus 16S metabarcoding) may become relevant later on.
- Microbial eukaryotes or single-celled protists are organisms that play a fundamental role in energy flow and the cycling of elements in the marine ecosystem. They occupy several different types of marine habitats. In northern latitudes where the presence of cyanobacterial lineages is diminished in marine waters, pico and nan sized marine microbial eukaryotes serve as the background primary producers of the system. Protist primary producers play an important role in structuring the microbial food web and in the transport of carbon to the deep ocean. They can be primary prodcuers, consumers, decomposers, and link trophic food chains in the marine food web. The Arctic is characterized by an extreme light climate, with the sun staying below the horizon for several months of the year. In line with this, light is known to be one of the main drivers for structuring microbial eukaryotic communities in the Arctic. A range of phylogenetically diverse protist groups are found in the Arctic more seasonally than based on any other environmental factor. Despite the abundance and importance of pico- (0.2-2µm) and nano- (2-20µm) sized eukaryotes, most protist research so far has focused on larger organisms that can be studied under a microscope. However, recent advances in molecular techniques have made it possible to address who is there? and what are they doing?, even for pico- and nano-sized protists, at the same time avoiding cumbersome microscopic analysis. Molecular methods are independent of morphology and instead use information from DNA sequences to identify organisms and classify them based on their evolutionary relationships. The 18S rDNA (the gene encoding the RNA component of the small ribosomal subunit) is a genetic marker that has been used successfully to characterize protist communities. Molecular tools can be used to identify the composition of microbial species in an environment (metabarcoding), to estimate species abundance (qPCR) or to study the gene diversity of the community (metagenomics). DNA and RNA markers give complimentary information; DNA provides estimates of species abundance and diversity, answering the question who is there, while RNA provides a picture of the active community. By specifically targeting protein-coding mRNA information can be obtained regarding what are they doing", giving a snapshot of the community at the time of sampling (e.g. metatranscriptomics). Furthermore, molecular genetic tools can be used to identify ecotypes endemic to the region of study (e.g. the Arctic) help in the recognition of taxa with specific biogeographic patterns and link distinct protist assemblages to environmental factors (e.g. water masses). The Isfjorden Adventfjorden station (IsA time series station) is particularly well suited for studying protist responses to such environmental changes. Located at the mouth of Adventfjorden within Isfjorden- one of the largest fjord systems of Spitsbergen, IsA is periodically influenced by influxes of Atlantic Water (AW) from the Wester Spitsbergen Current. During winter, Isfjorden is often dominated by cold and fresh arctic watermasses while in summer there is an observed dominance of warm and saline AW. However, great interannual variation can be seen. The IsA time series station (78°N and 15°32´E, 85m bottom depth) has been sampled weekly to monthly since 2011 by UNIS, and is still sampled on a montly basis. Sampling will continue as part of this PhD project. The station is used to study the baseline biodiversity of microbial eukaryotes and zooplankton, as well as interannual variations in organisms and environmental factors. For this PhD I will use previously acquired data, collected samples and new sampling to

investigate potential changes in biodiversity, community composition and function of microbial eukaryotes with respect to the changing climate in the Arctic. More specifically I aim to: 1) Temporally classify the diversity of eukaryotic protists at the IsA station over the past years (2011-2019) and possibly model future conditions, 2) Compare microbial eukaryotic communities from "model systems" representing a "native" seasonally ice-covered Arctic (Svalbard east coast) and a future "Atlantified" ice-free Arctic (Svalbard west coast), 3) Quantify seasonal and diurnal light harvesting (photosynthesis and rhodopsins-mediated energy capture) in protists from Svalbard and compare the results to similar measurements from a less extreme light regime (southern Norway).

- I´m mainly involved in RF3 with several more logsitically task as I´m employed as research engineer, but working aside with barcoding of sea ice meiofauna (with Rolf Gradinger, Bodil Bluhm) and metabarcoding of sediment trap data (Marit Reigstad, Yasemin Bodur).
- As part of my PhD project, I will be studying the seasonal and latitudinal patterns in primary production in the Northern Barents Sea. In an intensive field season that is currently taking place, I am assessing total particulate primary production, which fraction of this production is fueled by nitrate inputs to the system (new production), and its response to light intensity. I am also conducting a range of on board and land-based experimental work. By comparing the functioning of Arctic communities to those found in e.g. subarctic fjords, I hope to elucidate how the functioning of key processes such as bloom seeding and onset might change in the face of Atlantification. Further along the process and in collaboration with the Phytochange research group at the Alfred Wegener Institute, I will carry out mock community experiments to understand the mechanisms behind primary producers' responses to multiple climate change stressors.

**Motivation for applying** *

Describe briefly why you want to participate in the workshop.

- I am super interested in joining this workshop as it addresses an interesting topic for my future career development and will provide me the tools I need to carry out biodiversity studies with the the most used techniques today
- This workshop could give me new insights in how to analyse amplicon data, as I have used the qiime and qiime2 platform before. Additionally this workshop is a fantastic opportunity to meet other researchers in AeN that will produce this type of data and will do these types of analyses in different contexts and therefore a good way to potentially streamline all amplicon analyses within AeN.
- I will be using metabarcoding to identify the communities in my samples, as well as further analysis using association networks and multivariate statistical methods to analyse community composition and gradients in the communities.
- I will be using metabarcoding as a core part of my project, and this workshop seems to cover most (if not all) aspects of my analyses.
- organiser, learn DADA2+networks
- Because metabarcoding is central to my thesis.
- I wish to apply for this workshop, to achieve all the goals of my PhD. I wish to dig deeper into the molecular techniques and learn from people all over in this field. I also wish to improve my undertsanding in R, and wish to be able to translate more numbers into valid information. I believe that this workshop will give me a better understanding of the fields of my current study.
- I would like to participate to keep up with todays analyses of NGS data, to incooperate that in my side research task in Arven etter Nansen. However, Marit Reigstad recommened that I only may participate if there is free space and priority should have PhD, Post Docs and Researchers.
- Even though my sampling and analysis could not be considered metabarcoding per se, I do study how several environmental factors alter community composition and function in experimental settings. I would like to have the proper toolset to do so in accordance with the rest of the Nansen Legacy members.

**Position, affiliation and supervisor** *

- Postdoctoral fellow I University of Oslo, Department of Biosciences I Dag Hessen & Alexander Eiler
- postdoc, UiB, Gunnar Bratbak
- Master Student, EvoGen, Håvard Kauserud
- MSc student, EVOGENE, Håvard Kauserud
- Ass. professor UNIS
- Phd-student, UNIS/UiT, Anna Vader
- PhD student, UNIS, Anna Vader
- Research engineer, UiT, main head: Marit Reigstad
- PhD Candidate, UiT the Arctic University of Norway, main supervisor Prof. Rolf Gradinger.

**Role in Nansen Legacy** *

Describe in which reasearch focus (RF) and task you are involved.

- I am not involved in Nansen Legacy
- RF3, T3-1.1 Characterize microbial communities; T3-2.1 Describe seasonality in microbial community; T3-4.1 Microbial food web
- N/A
- N/A
- RF3-1,3-2,3-4
- RF3, task T4-4.1
- T3-2.2
- RF3
- My work is conducted within RF3 (The living Barents Sea). According to the Nansen Legacy implementation plan, my position is involved in tasks T3-1.1, 1.2, 1.3, 2.2, 3.1, 3.2, 4.1.

**Have you previously used R?** *

- I have a basic knowledge about using R, which currently I am applying to metagenomic data analysis
- Yes, but only occasionally for some specific applications.
- Yes.
- Yes.
- yes, briefly
- Have used it, but not much. Can do the most basic stuff.
- Yes
- yes
- I have used R to perform standard descriptive statistics, fit linear regression models and to carry out multivariant analysis of species abundance datasets (PCA, NMDS, cluster analysis).

Se nylige endringer i Nettskjema (v814_0rc1)