Final project report
Reporting template

Project acronym		CLIMARCTIC
Project title		Climate change impacts on Arctic soil and lake microbiomes
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	Entity (Company/organization)	Ghent University, Belgium
Project period (Start date – End date)		01/03/2017 – 30/11/2020
Project website, if applicable		www.climarctic.ugent.be

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List of partners involved in the project (company/organization and principal investigator). Please use partner numbers to specify the tasks, work packages and inputs of each partner in sections 4.3, 5 and 6.2 to 6.4.	Partner 1: Elie Verleyen, Ghent University, Belgium Partner 2: Mette M. Svenning, UiT the Arctic University of Norway Partner 3: Beat Frey, Swiss Federal Institute for Forest, Snow and Landscape Research, WSL Partner 4: Antonio Quesada, Universidad Autónoma de Madrid, Spain Partner 5: Ulf Karsten, University of Rostock, Germany

1. Short description for publicity

Nowhere is climate change more visible than in the Arctic, making it the most critical reference region for the detection and understanding of global change, and its effects on biodiversity and ecosystem functioning. The physical and chemical attributes of Arctic soils and lakes are very sensitive to climate change because of their close proximity to freezing, implying critical limits to snow cover, albedo, ice extent and phenology, and light availability. It follows that relatively small climatic changes can have profound impacts on Arctic ecosystems and their biota. This rapid response is also due to the fact that biogeochemical cycling and ecosystem functioning in Arctic environments are to a large extent controlled by microorganisms from all domains of life. The CLIMARCTIC project was aimed at studying the effects of climate change on the diversity and genetic functional attributes (nutrient and carbon cycling) of a high-Arctic terrestrial microbiome in soils, wetlands and lakes. To achieve this, we studied the diversity and functional genomic make-up of tundra and their substrate soils in two transects along a moisture gradient in Ny-Ålesund (Svalbard) using high-throughput DNA and RNA sequencing. This was combined with laboratory and field experiments and fossil DNA analysis to assess the effect of seasonal and long-term changes in temperature and precipitation on tundra and lake microbiomes. As such, CLIMARCTIC has developed a valuable platform to study the impact of future climate and environmental changes on high-Arctic tundra ecosystems.

Picture 1: Sampling of tundra communities and their underlying substrate soils in Ossian-Sarsfjellet (July 2017). The samples for DNA analysis were taken aseptically and immediately stored in liquid nitrogen to prevent DNA degradation.

Picture 2: Sampling of tundra in Knudsenheia (Ny-Ålesund) during winter (February 2019). After removal of the snow cover, soil cores were taken and immediately frozen in liquid nitrogen to prevent the degradation of RNA. Similar samples were also taken during spring and summer to study seasonal changes in the functional and taxonomic diversity and activity of microbial communities and their effect on the soil carbon cycle.

2. Summary

CLIMARCTIC studied the effects of climate change on the diversity and genetic functional attributes (nutrient and carbon cycling) of high-Arctic terrestrial microbiomes in soils, wetlands and lakes in two sites in Ny-Ålesund, Svalbard. To achieve this, a space-for-time substitution approach was used along gradients in soil moisture to examine the surface and subsoil microbial communities. This was combined with the installation of snow fences to experimentally increase snow cover in dry and wet tundra systems. A suite of environmental variables and ecosystem functions were analysed and combined with high-throughput amplicon sequencing, metagenomics and metatranscriptomics of contemporary and fossil environmental samples, as well as soils from controlled laboratory experiments. The key findings are:

i. Significant differences in gravimetric water, soil organic matter and nutrient contents were coincident with a shift in vegetation cover and species composition along the environmental gradients in the two sites. The potential enzyme activities varied significantly across the sites, as did the trophic interactions between the soil groups, but also with depth and soil moisture.

ii. High-throughput amplicon sequencing revealed that microbial communities were significantly different between the surface and sub-surface soils. Differences in microbial community structure in the surface soils were mainly related to variations in soil moisture, which was confirmed by a photosynthetic marker pigment based approach for the photoautotropic taxa. By contrast, soil organic matter, carbon and nutrient concentrations appeared to better explain differences in community structure in the sub-surface soils. Our study suggests that future changes in soil moisture will have a differential impact on surface versus sub-soil microbial community structure in Arctic tundra.

iii. The input of organic material with distinct chemical structures into high-Arctic soils has important implications for microbial community diversity and its functional feedback on soil C turnover. We assessed the effect of increased temperature and the addition of both labile and relatively stable fresh plant derived material on greenhouse gas production and bacterial and fungal community structure and activity. Elevated temperature promoted CO_2 release from the soils amended with both substrates, but a greater positive priming in warmer soils was observed with the addition of the more labile carbon source.

iv. Analysis of fossil DNA and pigments in radiocarbon dated sediment cores from Lake Sarsvatnet revealed the presence of mosses in the catchment area immediately after regional deglaciation. Dinoflagellates, diatoms and green algae dominated the phytoplankton following the retreat of glacier ice in the lake's catchment.

3. Objectives of the research

CLIMARCTIC was aimed at studying the effects of climate change on the diversity and genetic functional attributes (nutrient and carbon cycling) of high-Arctic terrestrial microbiomes in soils, wetlands and lakes. The combination of paleolimnological, field studies and laboratory experiments provided insights into the response of these poorly understood ecosystems to climate changes, and hence to assess their role as feed-back mechanisms in the climate system under future global changes.
The specific objectives of CLIMARCTIC were to:

(1) study the biodiversity and structure of dormant and active microbial communities and food webs in soils and lakes in the high-Arctic along environmental and temporal (diurnal and seasonal) gradients;
(2) assess the role of biodiversity and climate change on ecosystem functions such as carbon, nitrogen and phosphorus cycling in soils and lakes;
(3) study the ecophysiological and biogeochemical response of these communities to changes in temperature, water availability, growing season and light climate;
(4) quantify source-sink dynamics in nutrients, carbon and biota between soils and lakes, and study the connectivity between these ecosystems;
(5) study the rate of change and temporal (natural) variability in food web structure and the organic carbon, nitrogen and phosphorus levels in lake sediments and the soils in their

catchments on a decadal scale during the past 2000 years, with a particular focus on well-known climate anomalies in the Northern Hemisphere, and assess how these responses compare with changes observed along gradients in temperature and moisture availability in present-day communities.

4. Project activities and achievements

4.1. General description of activities over the duration of the project

The project was organized in four interrelated scientific work packages and a work package aimed at stakeholder involvement and integration (see 5 and 6). Field campaigns were organized for sampling tundra, soils and lake sediments along transects in (seasonal) environmental conditions in two sites in Ny-Ålesund (Svalbard), and to install permanent monitoring plots and snow fences in one site. The AWIPEV station provided logistical support during all field campaigns. All partners participated in the field campaigns.

WP1: Biodiversity and food web structure

This WP aimed at studying the biodiversity and food web structure in soils and lakes along environmental gradients by using a space-for-time substitution approach. This objective was addressed using two different approaches, namely (i) high-throughput amplicon sequencing of taxonomic markers genes in combination with the analysis of environmental properties, and (ii) studying the main components of the food webs using microscopy, pigment biomarkers and a stable isotope labelling experiment.

Task 1.1: High-throughput amplicon sequencing of taxonomic marker genes

A total of 108 samples were taken from biological soil crusts and their substrate soils along transects in water availability in the two field sites (Ossian-Sarsfjellet and Knudsenheia) during the 2017 summer campaign. The in situ samples were subsampled in the AWIPEV laboratory in Ny-Ålesund for the analysis of abiotic properties and DNA/RNA extraction. DNA and RNA extraction was performed in the laboratory of P2 in collaboration with P1 using the Qiagen RNeasy PowerSoil Total RNA and DNA elution kits on samples ground under liquid nitrogen in order to preserve the functional signal. Subsamples of the DNA extracts are being stored at -80 °C in the laboratory of P2, and another subsample was sent to the laboratory of P1 for amplicon library preparation in collaboration with all partners. A total of four primer sets were used to obtain amplicons of the 16S rRNA and 18S rRNA genes, and the ITS2 region for studying the bacterial and cyanobacterial, eukaryotic, and fungal diversity, respectively. In addition, pH, soil moisture, soil organic matter (SOM), carbon (C), nitrogen (N) and sulfur (S), total C/N, sand, silt and clay were measured by P5 and P3. These environmental data were combined with a vegetation survey of the sampling sites (P5). The amplicon sequencing datasets were processed using bioinformatics pipelines by P2 in collaboration with P1 based on Tytgat et al. (2016, submitted) using the PEAR (Zhang et al. 2014), USEARCH (Edgar 2010), Uchime (Edgar et al. 2011), UPARSE (Edgar 2013) and Mothur (Schloss et al. 2009) packages, and subsequently analysed using multivariate statistics in R. The results are currently being summarized in scientific publications.

Task 1.2: Food web structure

In this task, the composition and relative abundance of the community involved in the food web, grouped in primary producers and consumers (microfauna), was studied using microscopy (P4). This was combined with high performance liquid chromatography (HPLC) of the photosynthetic marker pigments (P1) following the protocols described in Tavernier et al. (2014). For ^{13}C and ^{15}N natural abundance analysis, samples were manually disaggregated, and microorganisms were sorted using a stereozoom microscope for $\delta^{13}C‰$ and $\delta^{15}N‰$ natural abundance signal analysis. Individuals of different trophic levels were manually separated by P4 under the microscope by microdissection and encapsulated in triplicates in zinc cases. Stable isotopes of carbon were also used as tracers of the food web, providing information about matter transfers within the community by an *in situ* experiment. The community was labeled with ^{13}C by exposing it under sunlight to $NaH^{13}CO_3$ which was

photoassimilated by the primary producers. After an incubation period of 24 h *in situ*, autotrophic organisms, mostly photoautotrophs, should have assimilated the isotope, and excess of unassimilated isotope was removed. Samples were obtained at time periods of 0, 24, 48 hours and 5, 6, 7 and 10 days. The different compartments of the community were encapsulated by P4 as described previously for natural abundance analysis.

WP2: Functional diversity in response to environmental variability

This WP was aimed at studying specific functional genes, the active organisms present and differential gene expression patterns in dry and wet tundra soils in response to environmental change using field and laboratory experiments.

Task 2.1: Functional diversity along environmental gradients

A set of functional genes involved in the nitrogen, phosphorus and carbon cycles (nifH, nirS, phoD, pmoA-681 and pmoA-682) were analysed and processed in the 108 samples using the procedures described in T.1.1 (all partners). This amplicon sequencing approach was combined with a metagenomics analysis of a selection of these samples (n=72; all partners). This metagenomics dataset was processed by P1 and P2 using the SqueezeMeta pipeline (Tamames and Puente-Sánchez, 2019). Quality control was assessed using FastQC (Andrews 2010). Adapter removal and quality trimming were done using Trimmomatic (Bolger et al. 2014) and SqueezeMeta was run in merge-mode to account for the large number of samples and data size. Assembly was performed using SPAdes (Bankevich et al. 2012) and gene prediction was done using Prodigal gene prediction software (Hyatt et al. 2010). Ribosomal RNA was predicted using Barrnap (Seemann 2013) and the RDP classifier (Wang et al. 2007) was used for classification. Functional assignment was done by homology searches against the eggNOG (Huerta-Cepas et al. 2016) and KEGG (Kanehisa and Goto 2000) databases. HMMER3 (Eddy 2009) was used for classification against the PFAM database (Finn et al 2014). The reads were mapped onto the assembly using Bowtie2 (Langmead and Salzberg 2012). Binning of contigs was done using Maxbin (Wu et al 2015) and Metabat2 (Kang et al 2015) and the binning results of both tools were merged using DAS Tool (Sieber et al 2008). Quality of the bins was checked using CheckM (Parks et al 2015).

Task 2.2: Functional gene activity along spatial and temporal scales in the field

For this task metatranscriptomics was combined with field and laboratory measurements of ecosystem functions. RNA was extracted from 108 samples from three seasons (spring, summer and winter) and processed as described in Task 2.1. In an additional selection of 36 samples, ribosomal RNA was not depleted to have an unbiased assessment of which microbial organisms are active during the respective seasons based on the 16S and 18S rRNA genes. Ribosomal RNA was predicted using Barrnap (Seemann 2014), and RDP classifier (Wang et al 2007) was used for classification. This approach was combined with gas flux measurements in the field by P3, and an analysis of enzyme activity (P5 and P3) as described in Pushkareva et al. (2020). In addition, long-term field experiments were initiated and aimed at assessing the response of tundra biomes on increased moisture availability and winter snow coverage. This was achieved by installing snow fences in a dry and a wet tundra site in Knudsenheia (all partners). Baseline samples from the start of this experiment are stored at the laboratory of P2, and nucleic acids were extracted and sequenced together with the transect samples. Nucleic acids are stored at -80 °C.

Task 2.3: Carbon cycling efficiency in response to environmental fluctuations in laboratory experiments

The effects of climate warming and plant input on the structure of the microbial communities breaking down soil organic matter in dry and wet tundra systems was studied using a laboratory experiment by P3 in collaboration with P2 and P1. The use of glycine (readily available substrate) and cellulose (more stable substrate) by prokaryotic and fungal communities was assessed in tundra soils incubated at 8 °C or 16 °C. A stable isotope probing (SIP) incubation experiment was conducted in microcosms prepared with 15 g (dry mass) of either dry or wet tundra soil with 30 mg of either glycine or cellulose substrate or without substrate (control). The microcosms were then placed into climate chambers either at 8 °C or at or 16 °C corresponding to soil mid-summer temperature and elevated temperature, respectively. The microcosms were incubated for 21 days with continuous monitoring and adjustment

of water content, and CO_2 and $^{\delta 13}CO_2$ gas fluxes were measured. DNA from ^{13}C-enriched samples (glycine or cellulose ^{13}C-enriched substrate) and unenriched samples (^{12}C-substrate treatments and control) were retrieved by ultracentrifugation and extracted using the procedures described in Zumsteg et al. (2013) and Rime et al. (2016). PCR amplification of the 16S rRNA gene (region V3–V4) and fungal ribosomal internal transcribed spacers (region ITS2) was performed with both light and heavy fractions and the products were analysed using Illumina MiSeq.

WP3: Nitrogen, phosphorus and carbon cycling in response to climate change

Task 3.1: Nutrient concentrations and their different sources

In addition to the environmental properties analysed in T.1.1 also soil NO_3^-, NH_4^+, P_H_20, P_$NaHCO_3$, P_labile, total carbon and total nitrogen contents were analysed by P5 in a selection of 36 samples using the procedures described in Kern et al. (2019). In parallel, and for the first time in the Arctic, a deep chemical profiling of soil organic matter characterization was undertaken on these samples (Pushkareva et al. 2020).

Task 3.2: Carbon cycling in natural communities

Main processes involved in the C-cycling (respiration and CH_4 production) were studied over seasonal and daily cycles. Gas (CO_2 and CH_4) flux measurements were conducted by P3 during the four seasons using a closed chamber approach along the environmental gradients as described in T.2.2 following the procedures described in Frey et al. (2011) and Hartmann et al. (2014).

Task 3.3. Nutrient conversion rates

This task was aimed at studying the nutrient conversion rates in the dry and wet tundra systems by P4. The nitrogen mineralization rates in the soils were analysed using incubation experiments of peat cores as described in Bayley et al. (2005). The different elements of the N cycle were investigated in the different ecosystems: N_2 fixation by diazotrophs (both photosynthetic and heterotrophs) by acetylene reduction assay and ^{15}N assimilation. Nitrification and denitrification were estimated using a standard acetylene inhibition process as natural and potential activities. N_2O was measured using the closed chamber approach as described in task 3.2.

WP4: Past and recent ecosystem changes

This WP was aimed at assessing the food web structure in lakes in response to past climate anomalies, which will provide natural background levels of the responsiveness and resilience of these ecosystems and the soils in their catchment area to climate change (P1). Sediment cores were taken using a combination of a gravity and Livingston corer at the a depth of 16 m in Sarsvatnet (Ossian Sarsfjellet).

Task 4.1: Sedimentology and sediment chronology

The cores were visually described in the field and transported frozen to Ghent University for subsampling. Subsamples were taken for ^{14}C dating and the measurement of sedimentological properties as described in Tavernier et al. (2014).

Task 4.2: Temporal trends in food web structure

A protocol for the extraction and processing of aDNA from the sediment cores was developed at the laboratory of P1 (De Maeyer et al. in prep.). DNA was extracted from sub-samples using the procedures described in T.1.1. and amplicons of the 18S rRNA gene were sequenced on an Illumina MiSeq platform. Fossil pigments were analysed by P1 as described in T.1.2.

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4.2. Table of deliverables

Deliverable and Milestone Name			Lead partner (country and designation)	Date of delivery (mm/yyyy)		Comments
				Initially planned	Delivered	
Work Package	***Deliverable or Milestone***	***Full Name***				
WP5	D1	Contributions to working groups of the Arctic Council	All partners	03/2017	03/2017 and ongoing	
WP5	D2	Information delivery to the Norwegian Environment Agency and Svalbard integrated Arctic Earth Observing System	All partners	03/2017	03/2017 and ongoing	
WP5	D3	Input in policy support	All partners	03/2017	03/2017 and ongoing	
WP5	D4	Interaction with the International Arctic Science Committee and the European Polar Board	All partners	03/2017	03/2017 and ongoing	
WP5	D5	Interaction with working groups of EU-PolarNet	All partners	03/2017	03/2017 and ongoing	
WP5	D6 M1	Website with information for scientists and the general public	P1 in collaboration with all partners	06/2017	04/2017	
WP5	D7	Blogs of the field campaigns	P1 in collaboration with all partners	09/2017	07/2017 created	
WP5	D8	Field reports with information for stakeholders	All partners	10/2017	08/2017, 10/2018, 05/2019	
WP4	D9	Dated lake sediment cores	P1	03/2018	04/2020	The sediment cores were taken in July 2018 and sampled for multiproxy analysis and dating in September 2019
WP5	D10	Intermediate report 1	All partners	03/2018	03/2018	

Deliverable and Milestone Name			Lead partner (country and designation)	Date of delivery (mm/yyyy)		Comments
				Initially planned	Delivered	
WP5 WP3	D11 M8	Scientific publication	All partners, led by P5	11/2018	09/2019	A first publication describing the study sites included measurements of the abiotic properties of the soils
WP1	D12 M2, M12	Biodiversity data of microbial communities in soils and lakes based on amplicon sequencing	All partners	03/2019	03/2019	Publications are being written, but delayed due to COVID-19
WP2	D13 M2, M4, M5,M14, M15	Data on active and dormant community members over a seasonal cycle	All partners, led by P2	03/2019	08/2020	The winter campaign was organized in early February 2019 in order to prevent plot disturbance for the spring campaign.
WP3 (WP2)	D14 M13 M14, M15	Data on carbon cycling in response to changes in temperature and snow cover	P5, P3, P2	03/2019	09/2019	An autumn campaign was organized in 2019 so data are available for all four seasons
WP1	D15 M13	Data on food web structure of lakes and soils	P3, P5, P1, P4	03/2019	06/2020	HPLC data were delivered in time, but due to a changes in personnel in the lab of P4, the stable isotope experiment was delayed. The paper on food web structure is in preparation
WP2	D16 M2, M4, M5 M14, M15	Data on functional genes in response to changes in temperature, growth season and moisture	All partners, led by P2	03/2019	06/2020	Spring and summer samples were collected during the first 2 campaigns. The winter campaign took place in February 2019. The RNA sequencing was delayed due to COVID-19. The data are being processed
WP3 (WP2)	D17 M10	Data on nutrient and carbon concentrations and cycling	All partners, led by P5	03/2019	06/2020	cf. D15 and M13

Deliverable and Milestone Name			Lead partner (country and designation)	Date of delivery (mm/yyyy)		Comments
				Initially planned	Delivered	
WP4	D18 M11	Data on past turnover in community structure in response to past climate anomalies	P1	03/2019	04/2020	cf. D9
WP1	D19 M2, M12	Data on shifts in community structure along environmental gradients	P4, P1, P2, P3, P5	03/2019	03/2019	The data are available and the publications are being written
WP1	D20 M2, M6, M13	Information on nutrient and carbon cycling in the food webs	P3, P5, P1, P4	03/2019	06/2020	cf. D15 and M13
WP5	D21 M9	Intermediate report 2	All partners	03/2019	03/2019	
WP2	D22 M2, M4, M5 M14, M15	List of genes which are up and downregulated over a seasonal cycle	All partners, led by P2	03/2019	06/2020	cf. D16
WP5	D23 M6, M11, M12, M13	Scientific publications	All partners	12/2019	Not yet delivered - ongoing	All publications are being written (see separate excel table)
WP5	D24 M16	Biodiversity data stored in public databases	All partners, led by P1	03/2020	Not yet delivered - ongoing	Sequencing data will be made available after the publication of the respective papers
WP5	D25 M17	Final report	All partners, led by P1	03/2020	11/2020	
WP5	D26 M18	Information booklet for the general public	All partners, led by P1	03/2020	Not yet delivered - ongoing	This booklet will be compiled after all the data are published in international papers (see also 4.5)
WP5	D27 M14, M15	Scientific publications	All partners	03/2020	Not yet delivered - ongoing	cf. D23

4.3. Scientific outcomes

WP1: Biodiversity and food web structure

Task 1.1: Marker-gene based biodiversity assessment (all partners)

Significant differences in gravimetric water, soil organic matter and nutrient contents were observed along both moisture gradients. These differences were coincident with a shift in vegetation cover and species composition (Fig.1; Kern et al. 2019). While chloro- and cyanolichens were abundant at the drier sites, mosses dominated the wetter habitats and vascular plants the intermediate plots. Cryptogamic covers generally dominated with a maximum areal coverage up to 70%. Multivariate analysis revealed that soil moisture showed the strongest relation between patterns in vegetation functional groups, together with NH_4^+–N and pH.

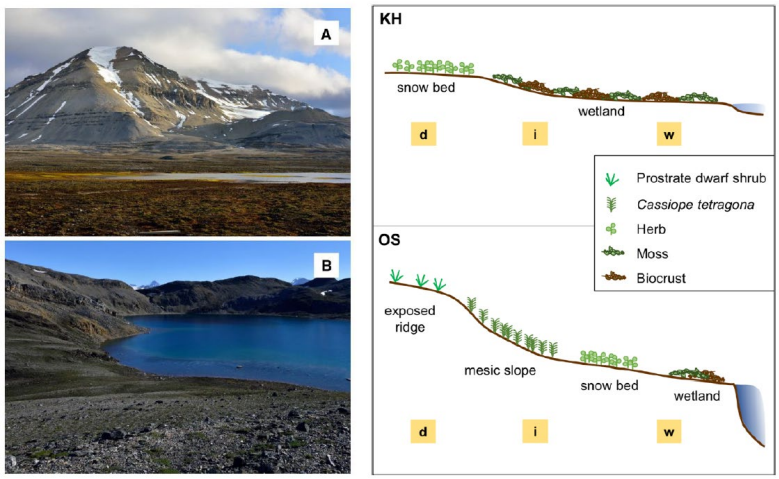

Fig.1: Pictures of the two study sites in Knudsenheia (KH, top) and Ossian-Sarsfjellet (OS, bottom) and a schematic diagram of the vegetation toposequences along the soil moisture gradients (d=dry, i=intermediate, w=wet) in both sites (Kern et al. 2019).

High-throughput amplicon sequencing of taxonomic markers genes in combination with the analysis of environmental properties in the 108 samples revealed that the microbial communities were significantly different between the top and sub-surface soils (Fig.2; De Maeyer & Seppey et al., in prep.). Proteobacteria was the dominant phylum in both sites and both top and subsoil samples across all moisture levels. Nevertheless, they were not the absolute dominant phylum in the subsoil samples, where the proportion of (non-phototrophic) Chloroflexi and Gemmatimonadetes increased. Cyanobacteria increased in the topsoils from dry to wet conditions, with an increasing amount of Nostocales apparent in the intermediate and wet samples. Chlorophyta was the most abundant eukaryote phylum, especially dominating the dry top soils from Knudsenheia. Ciliophora was the second most abundant group in the top soils, and most prominent in the subsoil samples. Their relative abundance increased from the dry to the wet top sites, but showed no obvious change in the deep samples. Cercozoan abundance and diversity increased from dry to wet, in both the top as well as and even more pronounced in the deep samples. A similar pattern was observed in the superphylum Stramenopiles, which was virtually absent in the dry top samples (and particularly diatoms), while chrysophytes were more widespread. The relative abundance of Basidiomycota approximately halves in relation to moisture and an opposite pattern can be observed for the Ascomycota, albeit much less pronounced.

Ordination and variation partitioning analyses confirmed that relative humidity was the most important factor in structuring the surface communities of Bacteria and Eukaryotes, and significantly explained respectively 14.2 and 13.9% of the variation in their composition. Soil texture and total carbon significantly explained variation in the community structure of Eukaryotes present in the soil surface, albeit to a lower degree than relative humidity. By contrast, total carbon and nitrogen concentration and the carbon-nitrogen ratio were generally more important in explaining variation in the sub-soil bacterial community structure. The eukaryotic community structure in the sub-soils was also significantly explained by total carbon and nitrogen concentration, although to a lesser extent compared to that of Bacteria.

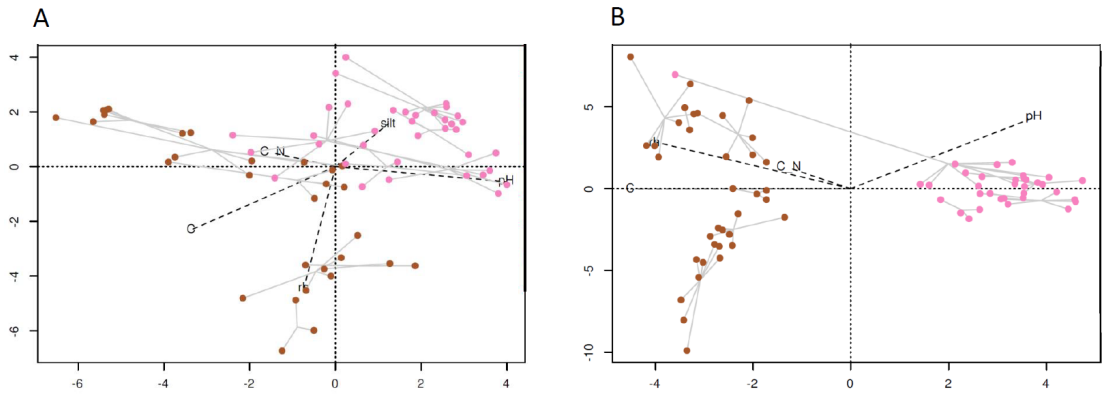

Fig.2: Parsimonious Redundancy analysis of the eukaryotic (A) and bacterial (B) communities in the surface (brown) and sub-surface soil (pink) layers. Variables significantly explaining the variation in community structure are indicated with striped lines.

Task 1.2: Food web structure (P4, P1)

High performance liquid chromatography (P1) revealed that the pigment concentration (both chlorophylls and carotenoids) was significantly higher in the wet sites compared with the dry tundra systems. In all plots, pigments related to land plants and green algae (chlorophyll b, zeaxanthin, lutein) dominated the profiles. Pigments related to cyanobacteria (myxoxanthophyll, echinenone) were generally more dominant in the wet sites. Marker pigments produced by diatoms and chrysophytes (fucoxanthin and diadino/diatoxanthin) were present at low concentrations, yet generally more abundant in the wet sites.

According to $\delta^{13}C‰$ and $\delta^{15}N‰$ natural abundance analysis of the studied groups and their ecology, trophic categories were established. Therefore, primary producers and the consumers category were defined. These communities varied according to the established trophic humidity gradient, where sample K1 was the driest and K3 the most humid.

The biodiversity in K1 was low and mainly composed of strictly terrestrial meiofauna (mites and collembola), while nematodes were occasionally present. The primary producers included cyanobacteria, green algae and mosses. For K3, primary producers included cyanobacteria, green algae and mosses, while the consumers consisted of tardigrades, rotifers and nematodes. The primary producers were substantially more abundant and more diverse in K3 compared with K1. The microscopy based identifications was corroborated by 16S and 18S rRNA metagenomic analyses. The green algae were composed of Ulvophyceae, Chlorophyceae and Trebouxiophyceae genera, while cyanobacteria belonged to the Nostocales (also Leptolyngbyales and Gloeobacterales). The 18S revealed the presence of Eutardigrada (for tardigrades), Bdelloidea (for rotifers), and Chromadorea and Dorylaimia (for nematodes).

The interpretation of the stable isotope data in R revealed important trophic interactions between cyanobacteria and consumers in K1, while green algae appeared to have a stronger trophic relationship with the consumers in K3. The circulation of C through different trophic levels was determined from changes in $^{13}C/^{12}C$ signatures through the food web at different time points. These analyses could not be carried out in the K1 soil samples because the abundance of primary producers and consumers was very small, so insufficient biomass was present. For K3, cyanobacteria and green algae showed similar behaviour, incorporating the labelled inorganic carbon during the first 48 h after incubation. The mosses incorporated this carbon more slowly, with the highest value of ^{13}C after 5 days. Rotifers and nematodes were the first consumers that incorporated the labelled inorganic carbon. Tardigrades showed a gradual enrichment of ^{13}C since the beginning of incubation, reaching the highest $\delta^{13}C‰$ at 7 days, and hence becoming the last organisms to assimilate the added inorganic carbon.

WP2: Functional diversity in response to environmental variability

Task 2.1: Functional diversity along temporal and environmental gradients (all partners)

The functional genes involved in the nitrogen, phosphorous and carbon cycles (nifH, nirS, phoD, pmoA-661 and pmoA-682) were analysed using MiSeq amplicon sequencing. These data are being processed using the pipelines used in Task 1.1. Biodiversity of methane oxidising bacteria (MOB) was obtained based on the primer pair A189f/mb661r for the *pmoA* gene, with PCR products for 64 of 108 samples. A cut-off value of 86 % for the *pmoA* gene sequence was used for the phylogeny (Wen et al., 2016). The A189f/A682r primer pair did not give results. We identified a dominance of atmospheric MOBs that have been identified in similar environments and is suggested to be widely distributed. The upland soil cluster of gammaproteobacteria (USC-gamma) dominated at both Knudsenheia and Ossian-Sarsfjellet while the highest abundance of alphaproteobacteria upland soil cluster (USC alpha) was found in the top layer at the KH dry site. MOB OTUs belonging to *Methylobacter*, *Methylovulum* and *Methylocystis* were also identified, but in lower abundances than the upland soil clusters.

The metagenomics datasets is being processed using bioinformatics pipelines. Due to measures taken to reduce the spread of COVID-19 (see also 4.5), the Illumina NovaSeq sequencing (also for Task 2.2) was significantly delayed. The dataset is 1,273,504 Mbp in size (resulting in a total of 4,2 G reads) and preliminary quality control revealed an average Phred score of 36 (94.36% of basepairs have a Phred score > 30). On average, samples have 58.6 ± 34.9 M reads, while one sample did not have sufficient reads.

Task 2.2: Functional genes along spatial and temporal scales in the field (P1, P3, P4, P5)

In total, 108 rRNA-depleted samples were submitted for library prep, of which 92 yielded sufficient RNA to be sequenced. One sample with a relatively low amount of reads (merely 1% of the other samples) was additionally removed after preliminary quality control. On average, Phred scores are 36 ± 0.14, implying that the need for trimming the reads is minimal. Per sample, there are on average 54.7 ± 1.5 M reads, with a total of 5.0 G reads (1,508,386 Mbp). Preliminary assembly tests resulted in contigs of 249-472 bp (N50 for the Spades and Megahit assemblers, respectively) long (with a maximum length of 12,185 bp). Taxonomic classification based on functional reads BLASTed against the NCBI nr database showed that Actinobacteria, Firmicutes, Proteobacteria and Acidobacteria were the most abundant prokaryotes. Archaea were recovered as well, with Thaumarchaeota being the most abundant group, although Euryarchaeota (i.a. Methanomicrobia) and several Asgardarcheaota representatives were also recovered. Streptophyta, Chlorophyta, Arthropoda, Nematoda and unclassified Chordata were the most abundant non-Fungi eukaryotes, while Ascomycota were the most important Fungi. Transcripts related to photosystem II P680 reaction center D1 protein were the most frequently recovered. Other abundant transcripts include methane/ammonia monooxygenase subunit C, hydrogenase large subunit and cold shock protein (beta-ribbon, CspA family). Ribulose-bisphosphate carboxylase large chain, too, was among the ten most abundant transcripts.

Task 2.3: Carbon cycling efficiency in response to environmental fluctuations in laboratory experiments (P3, P2, P1)

By testing the microbial utilization of glycine, a readily available substrate, and cellulose, a more stable C substrate, the SIP-DNA experiment demonstrated that the input of organic material with distinct chemical structures into high-Arctic soils has important implications for microbial community diversity and its functional feedback on soil C turnover (Frossard et al. subm.). Elevated temperature promoted CO_2 release from the soils amended with both substrates, but a greater positive priming of SOM in warmer soils was observed subsequent to the addition of glycine (Fig.3). Soil moisture did not affect the fluxes of CO_2, although distinct microbial communities characterized the dry and the wet tundra soils. Only few prokaryotic taxa responded to glycine amendment, whereas a high proportion of responsive taxa utilized only cellulose. Prokaryotic and fungal taxa responding to the substrates were mainly abundant taxa known to be fast growers, r-strategists or taxonomically acknowledged to have a putative copiotrophic lifestyle.

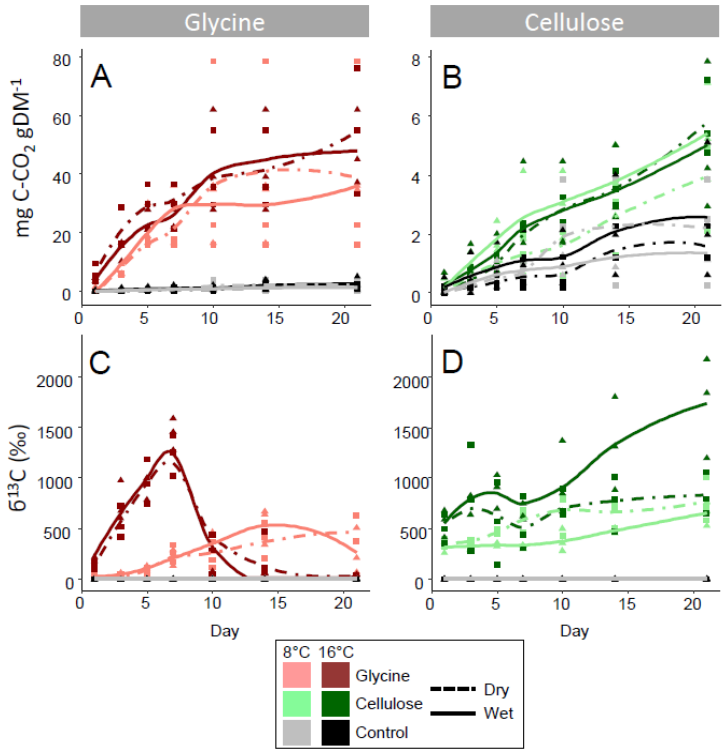

Fig.3: Cumulative fluxes of C-CO_2 (A and B) and $^{\delta 13}$C-CO_2 signatures (C and D) from the soils amended with either glycine, cellulose or no substrate (control) during the 21-day incubation experiment. Lines represent mean rates and individual measurements are shown as data points (n=3).

The output of this experiment helps to tackle the knowledge gap concerning the identification of microbial taxa actively participating in C cycling in high-Arctic tundra soils, a topic of high importance considering the increasing amount of C being made available through melting of permafrost soils and increased vegetation biomass.

WP3: Nitrogen, phosphorus and carbon cycling in response to climate change

Task 3.1: Nutrient concentrations and their different sources (P5)

The NH_4–N contents were always higher than those of NO_3–N (Kern et al. 2019). The NH_4–N values ranged along the water availability gradient between 30.31 and 49.17 mg kg^{-1} dry weight in KH and between 25.45 and 69.61 mg kg^{-1} dry weight in OS with a tendency of higher amounts in the dry subsites. The NO_3–N contents ranged from 16.24 to 48.65 mg kg^{-1} dry weight in the soil in KH and from 4.97 to 30.71 mg kg^{-1} dry weight in OS. The OS intermediate and wet plots exhibited with 46.78 and 10.34 mg kg^{-1} dry weight much lower values compared to the dry plots (23.62 mg kg^{-1} dry weight). In contrast to both nitrogen compounds, P-labile contents were always much lower with values between 3.02 and 4.97 mg kg^{-1} dry weight in KH, and between 1.82 and 2.59 mg kg^{-1} dry weight in OS. The C/N ratio ranged between 16 and 20 across all samples, thereby indicating clear N limitation at both study sites. The available P contents are also very low compared to other Arctic soils such as in Alaska or Canada, but there is still a general lack of knowledge on biogeochemical cycling and budgets of P in Arctic soils.

Task 3.2: Carbon cycling in natural communities (P5, P3, P2)

Gas fluxes of CO_2 and CH_4 at four different seasons along the soil moisture gradient at both Knudsenheia and Ossian-Sarsfjellet sites showed highly contrasting seasonal patterns.

While net primary production showed highest rates in summer, we could not detect any rates in winter. CO_2 fluxes in summer decreased along the gradient, with the dry soils being a source and the wet sites being a sink. In summer and spring, the dry soils were a sink for CH_4 whereas wet soils appeared to be a source in both sites. In autumn, CH_4 was emitted at both sites.

A chemical profiling of soil organic matter (SOM) was undertaken in soil samples from both study sites and two different soil layers (0-1 and 5-10 cm depth) along the moisture gradient using pyrolysis-field ionization mass spectrometry (Py-FIMS). The data demonstrate that SOM in both sites was dominated by lipids/sterols, alkylaromatics and phenols/lignin monomers. Dehydroergosterol prevailed in a majority of the samples. Biocrusts samples in both sites had higher total ion intensity (TII) and volatile matter (VM), but lower lipid and alkylaromatics contents (Pushkareva et al. 2020).

Task 3.3. Nutrient conversion rates (P4)

Samples from the two sampling sites and the three humidity conditions were taken and analysed in Ny-Ålesund. The samples were included in gas tight chambers and N fixation (acetylene reducing assay), and denitrification were analysed using ^{15}N stable isotopes. The potential denitrification at Knudsenheia depended directly upon the humidity in the soils. The areas described as dry showed a lower activity than the areas with a higher humidity. Short-term potential denitrification was higher at the medium humidity samples, but at the end of the experiment (72 h), the potential denitrification was maximum in the wet samples. However, at Ossian-Sarsfjellet potential denitrification was much lower and could not be detected. N_2-fixation showed a clear gradient in Knudsenheia in which significant differences were found among the different sampling sites. In that way N_2-fixation was almost 5 times higher in the wet soils compared with the dry ones, clearly reflects differences in the abundance N_2-fixers in response to different humidity conditions (see also T.1.1). However, values were lower at Ossian-Sarsfjellet and very close to the detection limit, and we observed no significant differences among the different humidity levels. These results indicate that N_2-fixation might be a relevant input of N in the high-Arctic soil ecosystem and that denitrification (measured as a potential activity) might be also a relevant player in the N balance in these soils. Liquid water availability seems to play a relevant role in relation to the N cycle in Arctic soils. In some places, N_2-fixation and potential denitrification were higher (Knudsenheia*)* in the more humid conditions, while in the drier sites (Ossian-Sarsfjellet) both N fixation and denitrification were lower and most likely balanced.

WP4: Past and recent ecosystem changes

Task 4.1: Sedimentology and sediment chronology (P1)

Two sediment cores with a length of 21 and 37 cm were retrieved from Lake Sarsvatnet at a water depth of circa 16 m. Radiocarbon dating revealed that the bottom of the first (C1) and second (C2) sediment core is older than 12700 and 18800 cal yr BP, respectively. The bottom sediments of both cores consist of glacial clay (Fig. 4) suggesting the presence of a glacier in the catchment area during the transition from the Last Glacial Maximum to the Holocene. Interpolation of radiocarbon dates in C2, in combination with the bottom ages of C1 suggests that the catchment area became ice-free around 13000 cal yr BP, which is in agreement with an unpublished study based on cosmogenic isotope dates of landforms (Grant 2016).

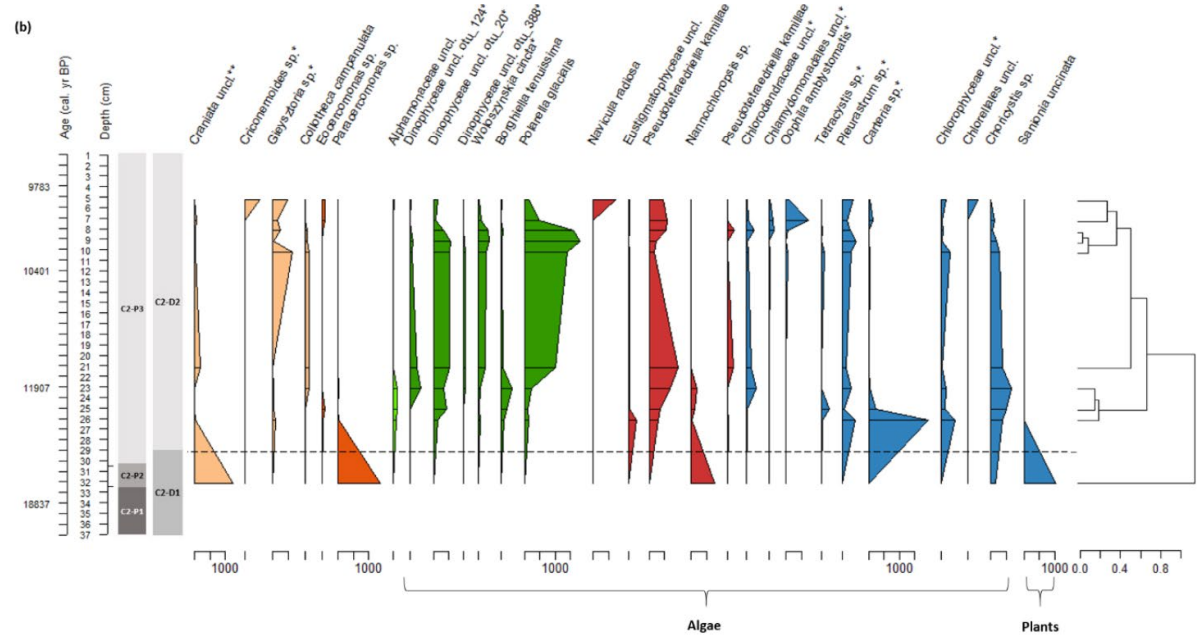

*Fig.4: Stratigraphic plots of the aDNA data in core C2 showing the OTUs belonging to Opisthokonta (light orange), Rhizaria (dark orange), Alveolata (greens), Stramenopiles (red) and Archaeplastida (blue). The plot shows the number of reads for each OTU after rarefaction. Pigment zones were indicated on the left for comparison. The classification OTUs indicated with an asterisk (*) is based on the NCBI database. **Had poor results when comparing to PR2 and NCBI database, and this OTU is presumably derived from polar bears based on alignments.*

Task 4.2: Temporal trends in food web structure (P1)

High performance liquid chromatography revealed that the concentrations of chlorophylls and carotenoids were extremely low in the glacial clay sections of both cores. Pigments derived from diatoms and dinoflagellates (diatoxanthin), and to a lesser extent from green algae and land plants (zeaxanthin, lutein), were the dominant markers in both cores. Fossil DNA analysis revealed the presence of OTUs belonging to the Opisthokonta, Rhizaria, Alveolata, Stramenopiles and Archaeplastida (Fig.4). In the latter taxon, the relatively high abundance of an OTU related to *Sanionia uncinata* in the glacial clay sediments suggests that this moss species colonized the catchment of the lake immediately after deglaciation of the region. An unclassified OTU related to a vertebrate and presumably to polar bears is also present in these bottom sediments.

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Pushkareva E., Eckhardt K-U., Hotter V., Frossard A., Leinweber P, Frey B., Karsten U. 2020. Chemical composition of soil organic matter and potential enzyme activity in the topsoil along a moisture gradient in the High Arctic (Svalbard). Geoderma 368: 114304.

Wen, X., Yang, S. Z., & Liebner, S. (2016). Evaluation and update of cutoff values for methanotrophic pmoA gene sequences. Archives of Microbiology, 198(7), 629-636. doi:10.1007/s00203-016-1222-8.

4.4. List of project meetings

Note that only general project meetings are included in the table below; (online) meetings for discussing specific results and the redaction of papers between partners, as well as joint laboratory work were not included.

Date	Place	Participating partners	Meeting title and object
15-16/03/2017	Ghent, Belgium	All partners	CLIMARCTIC kick-off meeting. Objective: select sampling sites, develop sampling strategy and protocol, plan first field campaign and general project management (e.g. hiring post-docs and budgetary issues)
20-22/06/2017	Ghent Belgium	Post-docs, PhD and MSc students from all partners	Sampling preparatory meeting. Objectives: explain protocols to students, pre-install and test snow fences, first aid course, and shooting and safety course for sampling in Svalbard
17/07/2017	Ny-Ålesund, Svalbard	PIs all partners	Pre-sampling campaign meeting. Objective: develop sampling strategy plan in order to ensure maximal collaboration and efficiency
31/10/2017	Tromsø, Norway	PI and postdoc P2, postdocs P1 and P4	Sample processing discussion. Objective: to select and finalise the DNA and RNA extraction protocol
28/03/2018	Rostock, Germany	PIs, postdocs PhD and MSc students all partners	Follow-up meeting. Objectives: to discuss and track the state of the deliverables and milestones, discuss the first data and to prepare the spring 2018 campaign
3-6/09/2018	Zürich, Switzerland	PIs, postdocs PhD and MSc students all partners	Follow-up meeting. Objectives: to discuss and track the state of the deliverables and milestones, discuss the publication strategy, discuss the ongoing lab work and preparation of the winter 2019 campaign; organisation of a bioinformatics workshop for postdocs and PhDs involved in the project
5-6/09/2019	Ghent, Belgium	PIs all partners, postdocs P1 and P2, PhD student P1; PI and post-doc P3 and post-doc P5 online due to field work/ongoing laboratory experiment	Follow-up meeting. Objectives: to discuss and track the state of the deliverables, milestones, and field work, and discuss the publication strategy and the ongoing lab work (metagenomics and –transcriptomics)

4.5 Follow up activities and plans for further exploitation of the results

- Due to the measures taken to reduce the spread of COVID-19 and sequencing companies prioritizing essential genetic analyses during the pandemic, the metagenomics and metatranscriptomics analyses were delayed. While these datasets were expected to be delivered by mid-March 2020, we only received the data in August 2020. This inevitably impacted the processing of these datasets, which requires a significant amount of computer time. The datasets are currently being analysed after the pre-processing (including quality control of the reads) that was finalized in September 2020. P1 and P2 will ensure the follow-up of these analyses, and the resulting scientific publications in collaboration with the other partners.
- The publications resulting from the work will be submitted to peer-reviewed international journals. Abstracts of several oral and poster presentations were submitted to be presented at different international symposia (i.a. ISME2020), but these were cancelled because of COVID-19. Hence, the results will be presented during future international symposia (e.g., ASSW2021, ISME 2022).
- During the first field campaign (July-August 2017), long-term monitoring plots and snow fences were installed. We foresee to take samples from these experiments during a follow-up project. Moreover, the samples obtained during the different field campaigns provide a very valuable platform for assessing the effect of future climate and environmental changes on tundra ecosystems. We will permanently store these samples and their DNA extracts (P2) and make these available for future research through continued collaborations between the partners of the project and other interested scientists.
- The datasets will be further used in review papers and student projects.

- Once all data are analysed and published, and field activities will resume in the Arctic, we aim to produce an information brochure about microbial communities in tundra ecosystems in response to climate change in the Arctic. The target groups for this information are the staff working in Ny-Ålesund, visitors in Ny-Ålesund and visiting tourists to Svalbard.
- We will continue to give lectures for school children and the general public as similarly done during the project. The CLIMARCTIC data will be used as case studies during lectures given by the partners in several bachelor and master programs at home universities.
- PhD students who have been working on the project are still engaged in activities organised by the Association of Polar Early Career Scientists (APECS). This engagement will remain after the end of the project.

5. Stakeholder engagement in the project

5.1 Before the project's start

AWIPEV station in Ny-Ålesund: consultation and involvement (all partners)

After approval of the project, we immediately started to identify a suitable and relevant region to conduct our fieldwork. P5 and P2 had ample experience with working in Ny-Ålesund. P5 therefore contacted the scientific coordinator (Dr. Ronald Neuber, AWI) of the German-French AWIPEV research station in Ny-Ålesund. This facilitated to efficiently (both in time and budget) organise the field campaigns and secured logistic support.

5.2 During the project

The Governor of Svalbard, Research in Svalbard (RiS) database and the Kings Bay science officer: consultation (all partners)

For each field campaign, the CLIMARCTIC team completed the applications for obtaining permits for sampling the terrestrial and lacustrine systems in Ny-Ålesund. To achieve this, the project was registered in the RiS database (https://www.researchinsvalbard.no) under the reference number 10774 and a summary of the planned field work was submitted prior to each sampling campaign. The Kings Bay science officer provided permits to install the permanent field experiments with snow fences in Knudsenheia.

AWIPEV station in Ny-Ålesund: consultation and involvement (all partners)

The logistic support of the field campaigns was provided by the German-French research infrastructure in Ny-Ålesund. The involvement of P5 in CLIMARCTIC was crucial in this respect. The field support included access to field sites by boat, the use of vehicles, bikes, field equipment and accommodation. Prior to the sampling campaigns the necessary forms were completed and reviewed by the AWIPEV station manager (Dr. Ronald Neuber, AWI, Germany).

Interaction with international programs and panels: involvement (P4)

The CLIMARCTIC project was represented by P4 who is actively involved in several international initiatives such as the European Polar Board (http://www.europeanpolarboard.org) and regularly participates to Arctic Council meetings. P4 informally introduced the CLIMARCTIC project at the annual meetings of IASC, and particularly at the Terrestrial Working group of IASC at its meeting in Helsinki (Finland). The H2020 CSA EU-PolarNet was informed about CLIMARCTIC initiative at the General Assembly in Madrid in 2019. Importantly, the participation of different countries in this project was presented as a merit in the Arctic Science Ministerial documents and meeting that was held in Berlin in 2018. It has been presented in the documents already submitted for the next ASM3 that will organized in Japan in May 2021. P4 has also included CLIMARCTIC in its report for the activities of the country (Spain) as an observer of the Arctic Council. Besides, CLIMARCTIC has been presented briefly at the Arctic Biodiversity Symposium in Rovaniemi (Finland) in 2018. The project's activities have been presented (not officially) at some working groups officers from the Arctic Council as Committee for the Arctic Flora and Fauna (CAFF)

Interaction with scientists: information and consultation (P1, P3, P5)
P1 organised a special session on microbial diversity in the Arctic during the Arctic Summit meeting in Prague (31/03 – 07/04/2017). P5 organised the 27th International Polar Conference in Rostock (25/03-29/03/2018) with various CLIMARCTIC contributions. P3 was leader of the session 'Functional ecology of polar microbial communities in a changing world' at the conference 'POLAR2018 – Where the Poles come together in Davos', 19th -23rd June 2018 with a contribution of CLIMARCTIC.

Interaction with the Svalbard Integrated Arctic Earth Observing System (SIOS): consultation and involvement (P1, P4, P5)
P1, P4 and P5 participated to a workshop regarding data publication and storage within SIOS (https://sios-svalbard.org) in Longyearbyen in August 2018. The data obtained in CLIMARCTIC will be made publically available after publication of the results in peer-reviewed journals (see also data management). The nature and position of the permanent field experiments were already communicated to SIOS. SIOS (M. Inger Jennings) provided logistic support to P1 for sending frozen samples to Belgium which were initially stored in their facilities in Longyearbyen.

Interaction with the Ny-Ålesund terrestrial ecology flagship program: collaboration (all partners)
P1 and P2 participated to a workshop for further developing and intensifying the activities within the Ny-Ålesund terrestrial ecology flagship program (9/11/2017). During this workshop a project proposal was developed for organising workshops and meetings. The project is funded by the Research Council of Norway. P1 and P2 are members of the scientific committee and work-package leaders (http://nysmac.npolar.no/research/flagships/terrestrial.html). P1, P4 and P5 attended the first workshop of the flagship program, which was organised in Ny-Ålesund (13-16/08/2018). P1 and P5 also attended the second workshop of the flagship program, which was organised in Longyearbyen (8-10/10/2019) during which the structure and content of the review paper were finalised. P1, P2, P4 and P5 have finally contributed to this review paper entitled '50 years of terrestrial and freshwater research in Ny-Ålesund, Svalbard: Current status and knowledge gaps' (Pedersen et al. in prep).

Czech polar research program: collaboration (P1)
Lake sediment coring was organized in collaboration with prof. Josef Elster (U. South Bohemia, Czech Republic) in July 2018 with the logistic support of the Czech polar research program. This collaboration resulted in a new project being funded by Ghent University (P1).

5.3 Foreseen after the project's end

Interaction with the Ny-Ålesund terrestrial ecology flagship program: collaboration
P1 and P2 will remain members of the scientific committee and participate in its meetings. This will ensure the valorisation of the results obtained within the CLIMARCTIC project. Moreover, this will lead to potentially future collaborations between scientists from other countries studying terrestrial ecosystems in Ny-Ålesund (see also below).

New research projects and initiatives resulting from the CLIMARCTIC project

- P1 obtained a grant funded by Ghent University to study the effect of bedrock characteristics and microtopography on the functioning and diversity of tundra biomes (2019-2023).
- P5 is involved in new DFG (German Research Council) projects (2021-2023) in Ny-Ålesund on biological soil crusts and soil protists.
- A former hired post-doc within the CLIMARCTIC project (with P3) will be a work-package leader in a newly funded BiodivErSA project (2021-2024) on the adaptation of soil microbial communities to climate change along large natural gradients from polar to hot desert environments (project 'GRADCATCH').

- P1 and P2 are WP leaders of the terrestrial ecology flagship project funded by the Research Council of Norway.
- A post-doc working on the project (P1) obtained EU-Interact funding for sampling soils in Svalbard in 2019 (BioSoCr project).
- P4 is involved in the Spanish Polar Program and the participation in CLIMARCTIC allowed him to facilitate a better exchange with the Svalbard community. Thanks to that, an official Memorandum of Understating (MoU) has been signed for exchange and collaboration with Japan, including the Japanese station in Ny-Ålesund. Two other MoUs are in preparation with Italy for collaboration in the Ny-Ålesund station and with Czech Republic for collaboration with their station in Longyearbyen. It is also relevant that the experience of P4 in CLIMARCTIC has promoted research on terrestrial Arctic topics in Spain and the number of applications for arctic terrestrial projects have increased by more than two fold in last 2 years.
- During the 2017 field campaign, samples were taken for the analysis of microplastics in Lake Sarsvatnet (P4). A publication in 'Science of the Total Environment' resulting from this work is already obtaining high attention and social and scientific impact as evidenced by the high number of downloads and citations in a very short period after publication. This, together with an additional manuscript being in the final phase of preparation, represents a relevant spin-off from the project.
- BelsPO funded MICROBIAN project (P1) to study the functional and taxonomic diversity in biological soil crusts in the Antarctic.

6. Dissemination of results

6.1 List of scientific publications

The publications are listed in the separate excel template.

6.2. Dissemination of results to scientists and scientific organisations (1-page max)

Organisation of special sessions during symposia and workshops

- Bioinformatics workshop (6/09/2018, Zürich, Switzerland). Workshop aimed at post-doctoral and pre-doctoral scientists.
- Special session on 'microbial diversity in the Arctic' (P1) during the Arctic Summit meeting in Prague (31/03 – 07/04/2017).
- Organisation of the 27th International Polar Conference in Rostock (25/03-29/03/2018) with various CLIMARCTIC contributions (P5).
- Special session 'Functional ecology of polar microbial communities in a changing world' (P3) at the conference 'POLAR2018 – Where the Poles come together' in Davos (19/06-23/06/2018) with a contribution of CLIMARCTIC.

Oral and poster contributions during conferences and symposia

- Frossard A., Hotter V., Kern R., Karsten U. and Frey B. Greenhouses gas exchanges and microbial activities in dry and wet high-arctic soils. Polar 2018 - A SCAR and IASC Conference, 18-23/06/2018, Davos, Switzerland. Oral
- Frossard A., Hotter V., Kern R., De Maeyer L., Tytgat B., Karsten U., Svenning M., Verleyen E. and Frey B. Microbial activities and fluxes of greenhouses gases in high-arctic tundra soils. 17th International Symposium on Microbial Ecology (ISME17), 12-17/08/2018, Leipzig, Germany. Oral
- Frossard A., Donhauser J., Niklaus P., Rime T. Frey B. Warming and reduction of precipitations affect the microbiome of recently deglaciated soils in the Swiss Alps. PAM 2017 - Polar and Alpine Microbiology Conference, 8-12/09/2017, Nuuk, Greenland. Poster

- Kern R., Hotter V., Karsten U., Climate Change Impacts on Arctic Soil and Lake Microbiomes. 2017. Forschungscamp (Interdisciplinary University-intern Conference) Rostock, Germany. Poster Karsten U. Poster
- Hotter V., Kern R., Frossard A., Karsten U. Vegetation coverage is dependent on moisture availability in the high-Arctic. German Phycological Conference, 11-14/03/2018, Berchtesgarden, Germany. Poster
- Hotter V., Kern R., Frossard A., Karsten U. Vegetation coverage is dependent on moisture availability in the high-Arctic. German Polar Conference, 25-29/03/2018, Rostock, Germany. Poster
- Aline Frossard, Lotte De Maeyer, Bjorn Tytgat, Mette Svenning, Elie Verleyen and Beat Frey. Microbial carbon utilization in dry and wet Arctic tundra soils under elevated temperature. Swiss Microbial Ecology: 30/01/2019-01/02/2019, Lausanne, Switzerland. Poster.
- Aline Frossard, Lotte De Maeyer, Bjorn Tytgat, Mette Svenning, Elie Verleyen and Beat Frey. Microbial carbon utilization in dry and wet Arctic tundra soils under elevated temperature. Final conference and strategic workshop, 12-14/11/2019, Brussels, Belgium. Poster

6.3 List of dissemination activities with stakeholders

- Description of the project for the Research in Svalbard database (see also https://www.researchinsvalbard.no). Summaries of the field reports have been uploaded after each field campaign.
- Project presentations for scientists working in Ny-Ålesund (July 2017 and July 2018) by P1.
- Verleyen E. 2018. CLIMARCTIC - Climate change impacts on Arctic soil and lake microbiomes. Ny-Ålesund newsletter n° 40: 10.

6.4 Dissemination of results to stakeholders (1-page max)

Dissemination of results to the general public (outreach) and education

- Article in newspaper (P5): Folgen des Klimawandels (NNN)
- Registration of the project, field campaigns and main results in the Research in Svalbard database (all partners)
- Lectures in public schools (e.g., Atheneum Voskenslaan Gent, Basisschool Kleine Prins Lede, …).
- So far, CLIMARCIC contributed to five MSc projects (P1: 3, P2:1, P5: 1) and three Ba projects (P1: 3).
- Contribution of P1 to the INTERACT publication 'Images of Arctic Science' (https://eu-interact.org/publication/images-of-arctic-science/).
- Publication in WSL Diagonal Magazine, "When soil in the tundra thaws, microorganisms heat up climate change".

7 Global Impact assessment indicators

7.1 Impact statement

CLIMARCTIC contributes to a more comprehensive understanding of an important permafrost ecosystem sensitive to climate change, and how annual seasonal changes can create perennial impacts in the Arctic. This knowledge is urgent for climate models as those used in the IPCC.

Dry and wet tundra were sampled along a moisture gradient in two sites in Svalbard. An extensive set of environmental parameters was measured in these samples, which allowed us to assess the effect of abiotic conditions on surface and sub-surface microbial community structure. This information is particularly lacking for the largely understudied dry tundra systems. A metagenomics approach

allowed to study differences in functional genes in these environments, which in combination with gas flux measurements, will result in a detailed assessment of genes and functions involved in the carbon and nutrient cycles.

A SIP-DNA laboratory experiment enabled to identify bacteria and fungi involved in carbon cycling in response to increases in temperature. Together with a soil metatranscriptomics study, this will allow to assess the active soil microbiome in dry and wet tundra systems.

The metatranscriptomics dataset also allow us to assess the functionality of the active soil microbiome at different seasons. This is to our knowledge one of the first studies providing detailed insights in the taxonomy and functioning of the soil microbial communities during the freezing and dark winter conditions in the high-Arctic.

This is the first occasion in which trophic interactions among different edaphic groups in high-Arctic soils are evaluated. This is expected to represent an important impact in the scientific knowledge, and would be a future reference for any soil ecological study in high-Arctic (and Antarctic) ecosystems.

Fossil DNA and photosynthetic pigment analyses in radiocarbon dated sediment cores were used to study successional patterns in lake and catchment communities since the deglaciation of the region. This provided insight into the responsiveness of these systems to environmental changes.

So far, the work resulted in 4 published and 2 submitted papers. The measures taken to reduce the spread of COVID-19, however, significantly delayed the receipt of the raw sequences from the sequencing centre for further collaborative bioinformatic downstream analyses of the metagenomics and metatranscriptomics data, and the redaction of the manuscripts resulting from this work. We envisage however that the data obtained will provide important new insights in the diversity and functioning of tundra ecosystems, which will be published in high impact peer reviewed journals and presented during international symposia.

Importantly, the project has developed a valuable platform to study the impact of future climate and environmental changes on high-Arctic tundra ecosystems. The publication of the data and accompanying metadata in public databases will facilitate the use and re-use of the results. The collaboration within the project has so far resulted in four follow-up projects. The project results have also been communicated to international organisations dealing with research in the Arctic and will be/were presented during international symposia.

7.2 Synthetic figures for the project publications (including interactions with stakeholders)

4 published papers, 2 submitted and 9 in preparation (listed in the publication list template, but not taken into account in the following table). **3** published papers with impact factor **>4** and 2 submitted papers to journals with an impact factor **>5** of which one in Nature Microbiology.

Analysis of the *project* publications:

Scientific Journal	*Number*	*Impact Factor (2018)*
Polar Biology	1	1.728
Geoderma	1	4.848
Frontiers in microbiology	1	4.235
Science of the Total Environment	1	6.551
Nature Microbiology	1	14.30
Environmental Microbiology	1	5.147

International dimension and multi-partnership for publications

		Number of publications
Multi-partner publications	**Peer-reviewed journals**	2 (and 1 submitted)
	Books or chapters in books	
	Communications (conferences)	6
Single-partner publications	**Peer-reviewed journals**	2 (and 1 submitted)
	Books or chapters in books	
	Communications (conferences)	3
Outreach initiatives including interactions with stakeholders	**Popularization articles**	2
	Popularization conferences	3
	Others	5

7.3. Other scientific outputs

	Number, years and comments (Actual or likely outputs)
International patents obtained	NA
International patents pending	NA
National patents obtained	NA
National patents pending	NA
Operating licences (obtained / transferred)	NA
Software and any other prototype	NA
Company creations or spin-offs	NA
New collaborative projects	6
Scientific symposiums	1 (+ 2 special sessions)
Others (please specify)	

7.4. Assessment and follow-up of personnel recruited on fixed-term contracts (excluding interns)

Identification			Before recruitment for the project			Recruitment for the project				After the project			
Surname and first name	Sex M/F	E-mail address	Last diploma obtained at time of recruitment	Country of studies	Prior professional experience, including post-docs (years)	Partner who hired the person (Organisation and Country)	Position in the project (1)	Duration of missions (months) (2)	End date of mission on project	Professional future (3)	Type of employer (4)	Type of employment (5)	Promotion of professional experience (6)
Ilse Daveloose	F	Ilse.daveloose@ugent.be	Graduaat Industriële Chemie	Belgium	18	UGent, Belgium	technician	7	12/2019	Fixed-term contract	university	technician	yes
Tine Verstraete	F	Tine.verstraete@ugent.be	Bsc Cel en Gentechnologie	Belgium	13	UGent, Belgium	technician	2.5	12/2019	Fixed-term contract	university	technician	yes
Luz Amadei Martínez	F	Luz.AmadeiMartinez@UGent.be	International Master of Science in Marine Biodiversity and Conservation	Spain	2	UGent, Belgium	Doctoral student	13.5	04/2020	fixed-term contract	university	Researcher	No promotion
Darja Belišová	F	Darja.Stojanovova@UGent.be	Master program in Biology	Czech Republic	2	UGent, Belgium	Doctoral student	6	12/2019	fixed-term contract	university	Researcher	No promotion
Aikaterini Pargana	F	Aikaterini.Pargana@UGent.be	Master on Biology - Animal Ecology	Sweden	2 years post-doc	UGent, Belgium	Post-doctoral	7	12/2019	post-doctoral position	university	Post-doctoral	No promotion
Aline Frossard	F	aline.frossard@wsl.ch	PhD	Switzerland	2	Swiss Federal Research Institute WSL	Post-doctoral	7	31.08.2018	Fixed-term contract	Public research	Researcher	Yes
Magdalene Adamczyk	F	magdalene.adamczyk@wsl.ch	PhD	Germany	0	Swiss Federal Research Institute WSL	Post-doctoral	17	29.02.2020	Post-doctoral position	Public research	Researcher	Yes
Christophe V.W. Seppey	M	cse017@uit.no cseppey@protonmail.ch	PhD	Switzerland	4	Prof. Mette M. Svenning, UiT The Arctic University of Norway	Post-doctoral	38	10.10.2020	post-doctoral	Research public institution	analyse	no promotion
Velazquez David	M	David.velazquez@awi.de	Ph. D	Spain	3 years posdoc	UAM, Spain	posdoctoral	24		Fixed-term contract	EPIC	Researcher/interantional technician	yes

Cavalcante Erika	F	Erika.cavalcante@gmail.com	Ph.D.	Brazil	2 years posdoc	UAM, Spain	Posdoctoral	6		Between jobs			
Kern, Ramona	F	Ramona.kern@uni-rostock.de	PhD 2010	Germany	7	University of Rostock	Postdoc	20.5	31th May 20	Postdoc	University	Researcher	yes
Pushkareva, Ekaterina	F	ekaterina.pushkareva@uni-rostock.de	PhD 2017	Czech Republic	2	University of Rostock	Postdoc	21.5	30th Nov. 20	Postdoc	University	Researcher	yes
Glaser, Karin	F	Karin.glaser@uni-rostock.de	PhD 2016	Germany	2	University of Rostock	Postdoc	1	1st Sep 18	Postdoc	University	Researcher	yes
Albrecht, Martin	M	Martin.albrecht@uni-rostock.de	PhD 2017	Germany	1	University of Rostock	Postdoc	1	1st Sep 18	Postdoc	University	Researcher	yes

7.5. Data Management and timeline for open access

The FAIR principles will be applied. The data will be structured in the DarwinCore Event format, and contain information about the species/OTUs, such as geographical coordinates, ecosystem type, date and time of collection, technical metadata (expedition, equipment used, name of the observer,...), and the abundance or number of individuals. When possible, the data will be combined with additional information, such as environmental measurements.

The nucleotide sequence data will be documented by formatting metadata in the Minimum Information about any (x) Sequence (MIxS) format. This (meta-)data standard was developed by the Genomics Standards Consortium, and has been adopted by the INSDC. Sequences alongside the MIxS information will be archived on INSDC. The sequences will be linked to species occurrences (i.e. collection place and date).

All genetic data are currently stored on the data servers of P1 and P2. After publication of the data in peer-reviewed journals, the genetic datasets will be stored in public databased such as the NCBI Short Read Archive (https://www.ncbi.nlm.nih.gov/sra). This is required by peer-reviewed journals and will ensure the data will be preserved on longer time scales (>10 years). In addition, the appropriate metadata will be stored in the open access database of the Svalbard Integrated Arctic Earth Observing Systems (https://sios-svalbard.org/metadata_search), which will facilitate the use and re-use of the datasets.