Title of Project: The Impact of Global Warming on the Carbon Cycle of Arctic Permafrost: An Experimental and Field Based Study

Institution: Princeton University

Date Received: Nov. 2010

Principal Investigator: Tullis C. Onstott

Project Results:

The proposed project objectives were the following:

1. Perform an ecosystem level study of the effect of global warming on Arctic permafrost by performing long-term, thawing experiments on well-characterized, intact cores of active layer and permafrost collected from a proposed high Arctic permafrost reference ecosystem site where CO_2 and CH_4 fluxes, temperatures, humidity, soil moisture and nutrients, microbial diversity and activity are being measured. We will perform the same measurements during the course of our experiments and compare with those measured at the "reference ecosystem site".
2. Characterize and compare the organic and inorganic composition of the active layer with that of permafrost before, during and after the long-term thawing experiments in order to identify the components of the permafrost organic C pool that are metabolized when the permafrost is thawed and the respiratory pathways utilized to degrade them.
3. Characterize the rates of production and consumption and the vertical flux of DOC, O_2, H_2, CO, CO_2 and CH_4 in the cores as a function of temperature, pCO_2, surface precipitation, sunlight and fluid flow and compare these results to field observations.
4. Characterize the regulatory and metabolic network of the active layer and permafrost microbial communities, using metatranscriptomic and metaproteomic techniques to determine which members and/or pathways are actively cycling carbon (e.g. aerobic heterotrophic respiration versus anaerobic fermentation and methanogenesis) as the permafrost is thawed. Compare these results with similar measurements performed at the reference ecosystem site.
5. Develop SLIMER, a Scintillator-Layered Microscope for Environmental Research, which maps the relationship between the metabolic network and the microbial community by using ^{14}C labeled substrates identified in the thawing experiments and the carbon trophic cascade during the course of the long-term thawing experiments.
6. Utilize recently developed Cavity Ringdown Spectroscopy (CRDS) technologies to characterize the isotopic systematics of CH_4 and CO_2 in the core thawing experiments and *in situ* at the reference ecosystem site and to relate these compositions to the source organic matter and the biodegradation processes.
7. Construct a 1D model that incorporates the vertical distribution of the physical, chemical and microbial properties of the active layer and permafrost and utilizes the results of the thawing experiments to constrain the carbon turnover rates and the DOC, CO_2 and CH_4 release rates as a function of temperature, organic C composition and water content.

Field Program - Utilizing the core acquisition, sealing and storage techniques developed in the first year of the project, forty, 1 meter long cores were collected from a 7 meter diameter ice-wedge polygon located near the McGill Arctic Research Station (depth to permafrost is 70 cm) in late April of 2011 on Axel Heiberg Island and transferred frozen to Princeton University. Twenty cores were delivered frozen to the University of Tennessee and twenty were kept frozen at Princeton for thawing experiments.

During a second expedition to Axel Heiberg Island vertical profiles of the pore water chemistry, the CO_2 and CH_4 concentrations, and the $\delta^{13}C$ of the CO_2 in the active layer for the same polygon were measured. In addition the diurnal variation in surface temperature and CO_2 and CH_4 fluxes were recorded during the summers of 2011 to 2013.

No uptake of atmospheric CO_2 was observed in the ice-wedge polygons. Instead *in situ* CO_2 flux emissions were 3.0±2.9 mmol CO_2 m^{-2} hr^{-1} in July of 2011 (12 to 13°C soil temperature at 0-5 cm depth, 3

locations within a polygon) and 1.5±0.4 mmol CO_2 m^{-2} hr^{-1} in July 2012 (7-11°C soil temperature at 0-5 cm depth). These polygons were found, however, to be net sinks of atmospheric CH_4 with an uptake flux of -400±100 nmol CH_4 m^{-2} hr^{-1} in July 2011 and -200±100 nmol CH_4 m^{-2} hr^{-1} in July 2012. Nearby polygons and wetland with higher vegetation cover areas directly adjacent to Colour Lake yielded CO_2 emissions as well ranging from 0.7 to 24 mmol CO_2 m^{-2} hr^{-1} in July 2010 and July 2011. The vegetated areas also exhibited atmospheric CH_4 uptake with fluxes of -520 to -800 nmol CH_4 m^{-2} hr^{-1} in July 2012 (Wilhelm et al., 2012; Allan et al., 2014; Bennet et al., 2012). The CO_2 flux from the vegetated wetland was ten times that of the polygons.

Microcosm Experiments - Between September 2011 and March 2013 a series of aerobic and anaerobic incubation experiments were carried out at Princeton University on active layer and permafrost soil, respectively. The experiments were run at 4°C and 15°C. Some were run under 4 amendment conditions (no amendment, 50-80% water saturation, 1mM of acetate and bicarbonate) in 24-hr darkness or under illumination. Some were run under saturation conditions ranging from 30% (close to in situ conditions) to 100%. All exhibited higher initial CO_2 and CH_4 fluxes during the first 2 days compared to the subsequent 42 days. CO_2 flux varied slightly across different mineral amendments. Active layer microcosms incubated under light produced CO_2 at the highest rate (0.2 – 0.6 μmol gm^{-1} day^{-1}) and that was followed by active layer and permafrost incubated in darkness (0.2 – 0.3 μmol gm^{-1} day^{-1} and 0.03 – 0.05 μmol gm^{-1} day^{-1}). CH_4 consumption was observed in all active layer soils with the highest rate (0.25 nmol gm^{-1} day^{-1}) recorded from non-amended replicates incubated in darkness and the lowest (0.04 nmol gm^{-1} day^{-1}) from acetate-amended replicates incubated in light. Permafrost soil supplemented with acetate and bicarbonate generated CH_4 at the faster rate (0.03 nmole gm^{-1} day^{-1}). Given the additional C substrates, the observed CH_4 production was low, yet IC analyses indicate that the acetate was consumed. Negligible N_2O was detected. IC analysis also indicated large amount of organic acids (at the scale of ppm) were produced by active layer under anaerobic conditions. $^{13}CH_4$ amended microcosms revealed that ~50% of the $^{13}CH_4$ was incorporated into biomass and ~50% was respired at $^{13}CO_2$ (Burton, 2013).

Stable isotope probe (SIP) incubation experiments were completed at McGill University using ^{13}C labeled carbon substrates for H_2/CO_2, methanol and acetate in samples which contained a significant proportion of methanogens species. These experiments revealed a significant uptake of CO_2 in the H_2/CO_2 incubation indicative of autotrophic (probably acetogenic) activity (Allen et al. 2013).

Long Term Intact Core Thawing Experiments - Core thawing experiments at Princeton University took place in a 4.5°C cold room. Eighteen cores of 1 m length were subdivided into control cores, cores thawed under saturated conditions, cores thawed under natural hydrological conditions from both the polygon interior and the polygon rim, and cores thawed in the dark. The first 14 weeks of core thawing simulated top-down spring thaw in the Arctic by progressively lowering -3°C ethanol on the outside of the cores until the soil below the permafrost table (70 cm) was thawed. During the remainder of the 18 months of thaw the cores were held at 4.5°C except for the control cores, with a permafrost table maintained at 70 cm and -1.5°C. Headspace gas samples and water and gas samples from 4 different depths in the cores were collected throughout the experiment and analyzed for pH, anions, cations, DIC, H_2, CH_4, CO, O_2, N_2 and CO_2, and $\delta^{13}C$ of CO_2. Cumulative emissions of CO_2 were tracked and examined for significant changes in rates of emission. Subsamples of sediment were collected at 4 depths over 18 months for DNA extraction and SOC characterization.

During the simulated spring thaw, cores were found to emit up to 1.4±0.6 mmol CO_2 m^{-2} h^{-1}, with emissions rates increasing during thaw and the majority of the CO_2 derived from aerobic respiration of carbon matching the SOC profile. In the absence of a CH_4 spike in the headspace, cores were found to initially emit 92±34 nmol CH_4 m^{-2} hr^{-1} before decreasing to 11±11 nmol CH_4 m^{-2} hr^{-1} by the end of thaw. The introduction of CH_4 into the headspace at atmospheric concentrations (1.8 ppm CH_4) from week 15 onwards immediately resulted in the uptake of CH_4 at a rate of 160±27 nmol CH_4 m^{-2} hr^{-1} in all treatments, including saturated soils. Pore gas measurements of depths immediately after thaw revealed the pooling of CH_4 in the subsurface at 65 cm and in the permafrost (400-500 nM CH4) which decreased (100-200 nM CH_4) without reaching the surface. Taken in conjunction with metagenomic data showing the highest proportion of methanotrophic sequences at the 5 cm and 35 cm depth, a microbial sink of both atmospheric and subsurface CH_4 is inferred from 0-35 cm. High SO_4^{2-} concentrations in the pore water (2-6 mM) also creates a system wherein sulfate-reducing bacteria are thermodynamically favored to

compete with methanogens for H_2 and acetate, providing a constraint on maximum methanogenic rates (Stackhouse et al. 2014).

For the next 10 weeks CO_2 emissions remained fairly stable among all treatments. At week 25 emissions rates began to significantly increase for the cores from the polygon edge and the cores in the dark treatment (reaching 4.1±3.1 mmol CO_2 m^{-2} h^{-1} and 3.3±1.3 mmol CO_2 m^{-2} h^{-1} respectively), followed 10 weeks later by the control, saturated, and *in situ* treatments (reaching 0.6±0.6 mmol CO_2 m^{-2} h^{-1}, 1.3±0.7 mmol CO_2 m^{-2} h^{-1}, and 1.5±0.6 mmol CO_2 m^{-2} h^{-1} respectively). Increases in emissions rates corresponded to the period of time during which soil compaction associated with thaw was most intense, with thaw slump ranging from 4-16% of total core length. Cumulative CO_2 emissions were highest from the edge cores at 179±11 mmol CO_2 and lowest in the control cores at 34±2 mmol CO_2, equivalent to a loss of 0.4-1.9% of the estimated 9.5 moles SOC in each core. Cores with thawed permafrost emitted from 170-520% as much CO_2 as the control treatment, with aerobic heterotrophy the predominant pathway (O_2 uptake/CO_2 emission ratio ranging from 1.0±0.1 (dark cores) to 1.4±0.1 (saturated cores)). Assuming a constant emission rate from the end of the experiment and a 4 month Arctic summer, SOC turnover times are approximated as 1278±1260 years for the control treatment, 531±285 years for the saturated treatment, 465±180 years for the *in situ* treatment, 174±132 years for the edge treatment, and 216±84 years for the dark treatment. Although no differences were observed between treatments, the overall $\delta^{13}C$ of CO_2 became progressively heavier during the course of the experiment, from -25±4‰ to -17±5‰.

CH_4 oxidation rates, similar to CO_2 emissions, were impacted by thaw slump and observed to increase by 59±10 nmol CH_4 m^{-2} hr^{-1} across all treatments during weeks 24 to 33. During the remainder of the experiment CH_4 oxidation rates decreased in all treatments to an end rate of 7±17 nmol CH_4 m^{-2} hr^{-1}. Decreases in CH_4 oxidation rates from headspace CH_4 co-occurred with the gradual replacement of methanotrophs of the genus *Methlyocella* (primary methanotroph at the start of the experiment) with methanotrophs from the classes *Methanomicrobia* and *Methanobacteria*, the increase in subsurface CH_4 concentrations at 35 cm (340±84 nM), the increase in acetate concentrations at 65 cm and the permafrost (up to 0.3 mM), and the loss of SO_4^{2-} to sulfate reduction across depths (2-4 mM).

Long-term CO_2 emissions derived from intact core thawing experiments are unique in that they do not follow the fast-pool/slow-pool SOC exponential degradation models used to calculate turnover times in microcosm studies. Rather than exhibiting a rapidly decreasing CO_2 emission rate within the first few weeks before shifting to a steady-state slow emission, core exhibited a continued growth in CO_2 emission over the course of 18 months. CO_2 emission rates of the core thawing experiment 1) are within the range observed in the field, 2) suggest that turnover times from intact systems are faster than those observed in microcosms, and 3) imply that the system is currently degrading SOC faster than the $\Delta^{14}C$ record indicates has historically been the case. Changes in CH_4 surface flux were likewise dependent upon changes over long periods of time in both the aerobic active layer and the anaerobic subsurface. Taken together this data provides critical constraints on CO_2 and CH_4 emissions and behaviors for climate models from an understudied mineral cryosol in the high Arctic.

SOC Characterization - C and N analyses of the 2011 cores revealed that the upper 10 cm of the active layer contained 1 to 6% wt. organic carbon whereas from 10 to 100 cm the active layer and permafrost contained ~1% organic carbon from this location. The C/N values decreased from 90 in the upper active layer to 15 to 18 in the lower active layer and permafrost as the total soil N remained constant. FT-ICR-MS and C K-edge XANES analyses reveal that this organic carbon is aliphatic rich, O-poor and H-rich consistent with a lipid dominated composition with only minor lignin content (Sanders et al., 2014). The 7-month long thawing experiment of the 2010 core revealed a relative decrease in the lipid content. The $\delta^{13}C$ of the organic carbon ranges between -26 to -27‰ VPDB and is uniform with depth (Stackhouse et al. 2014).

Neutral lipid extracts yield molecular weights that are consistent with the FT-ICR-MS of acetone extracts. ^{14}C analyses of the total organic carbon, neutral lipids and residue yield ages ranging from 4,000-6,000 years (Ziolkowski et al., 2014).

DNA Extraction Protocol Development - Total community genomic DNA (gDNA) was isolated in triplicate from one sample of the 2011 cores using commercially available kits (FastDNA SPIN, MoBio PowerSoil, MoBio PowerLyzer and Meta-G-Nome). The bacterial 16S rRNA v3 region was amplified directly from gDNA (single PCR) or re-amplified from a 16S rRNA fragment (double PCR) followed by 454 pyrosequencing. The most abundant phyla (Actinobacteria, Firmicutes, Gemmatimonadetes, Proteobacteria, Acidobacteria, and unclassified Bacteria) detected using different DNA extraction

methods were significantly different (P=0.003 for single; P=0.032 for double PCR amplification). FastDNA SPIN generated high amounts of good quality DNA with minimal error between triplicates compared to other DNA extraction methods. Taxonomic analysis based on the RDP naïve Bayesian rRNA Classifier showed similarities in bacterial community regardless of DNA extraction method and number of amplification steps. Phylogenetic analysis of 24 amplicon libraries indicated that sequences affiliated with 284 genera representing 16 phyla were detected at least once. Sequences with abundances >0.29 ± 0.22% were detected in 90% of the 24 DNA extraction and amplification combinations. For the 9 most consistent samples, a sequence only needed a relative abundance of 0.06±0.05% to be detected in all 9 samples. The bacterial community structure from the Meta-G-Nome extracts differed from other kits exhibiting higher proportions of easily lysed β- and γ-Proteobacteria and lower proportions of *Actinobacteria* and *Methylocystaceae* important in carbon cycling. The results indicated that gDNA yields differ between the extraction kits, but reproducible bacterial community structure analysis may be accomplished using gDNAs from the three bead-beating lysis extraction kits (FastDNA SPIN, MoBio PowerSoil, and MoBio PowerLyzer). The sequences from 24 amplicon libraries were annotated in MG-RAST and made public under accession numbers 4478917.3-4478923.3; 4478925.3-4478929.3; 4478944.3; 4479284.3; and 4479286.3-4479295.3 (Vishnivetskaya et al. 2014).

Metagenomics – The microbial community changes were monitored during 18 months of controlled thawing under varying light exposure and water saturation conditions. For metagenomic libraries the gDNA was extracted from 5 cm, 35 cm, 65 cm and 85 cm deep samples collected from 17 one-meter long intact cores consisting of active layer and permafrost. A total of seventy-six metagenomes spanning the five treatment conditions (in situ, saturated, dark, edge and control) and five time points (0, 1 week, 6, 12 and 18 months) were generated and raw metagenome sequences were uploaded to NCBI Sequence Read Archive under the accession number SRP047512. Permafrost thawing resulted in an increase in anaerobic fermenters and sulfate-reducing bacteria but not methanogens. Amplicons as well as both unassembled and assembled metagenome sequences from thawing experiment were uploaded to MG-RAST and represent a set of 219 sequencing libraries. Forty-three assembled metagenomes from t=0, 1 week and 6 months thawing were uploaded to IMG website.

Analysis of amplicon and metagenome libraries for 16 (4 depths from 4 cores) samples from t=0 showed that microbial community structure varied considerably with depth displaying highest diversity in the uppermost layer. As estimated by Illumina amplicon, 454 shotgun and Illumina shotgun sequencing the phylum Actinobacteria dominated in permafrost ranging from 43% to 57% of the population. Actinobacteria were also a significant component of the active layer ranging from 18% to 28% of the population. Metagenome data showed that metabolic activity also varied with depth, and statistically significant differences were detected in nitrogen metabolism and carbohydrate utilization. Using these Illumina metagenome sequences a few pangenomes belonging to uncultured representatives from phyla TM7, Bacteroidetes, Chloroflexi, Firmicutes and domain Bacteria were reconstructed.

Three samples representing active layer, permafrost and mix of active layer and permafrost were used to setup microaerophilic microcosms at room temperature using selected media, such as 1/10 R2A, Glucose, TSB, Potato-dextrose. DNA was isolated using FastDNA SPIN kit. Metagenome sequences (both unassembled and assembled) were uploaded to MG-RAST and annotated representing 18 libraries.

Metaproteomics - Metaproteomics of all three pristine core samples from 2010 yielded very low amounts of extractable proteins, suggesting low microbial biomass, relative high percentage of dormant cells and/or lack of relevant annotated metagenome information for protein identification. Soil microcosms were set up using the same soil samples, subjected to different nutritional amendments at 10^{o}C. Little activity was noted in microcosm that received glucose as nutrition. Resultant protein datasets were initially matched with the Eureka metagenome and the proteome profile indicated presence of stress responsive proteins (e.g. DnaK, GroEL) and proteins essential for energy production and survival under carbon starvation such as F0F1 ATP synthase and acyl-CoA dehydrogenase. These proteins were annotated as belonging to genera *Bradyrhizobium, Sphingomonas, Lysinibacillus* and *Methylophilaceae.* The results were similar to those obtained by 454 pyrosequencing of the same samples and indicate low microbial activity in the native permafrost soils.

A subset of active layer samples was incubated with acetate at room temperature for 6 weeks, with unamended microcosm serving as control. The samples were analyzed via mass spectrometry and resulting datasets were matched with 1) the Eureka metagenome, 2) a selectively assembled database containing all sequenced psychrophiles, and 3) a selectively assembled database of microorganisms known for anaerobic oxidation of CH_4 (AOM isolates). The control sample yielded little amounts of protein

with few matches to any of the three databases. The acetate amended sample exhibited low matches to Eureka metagenome, but led to identifications of 163 proteins using psychrophile database and 170 proteins with AOM isolates database. Resulting datasets from both databases were populated largely by proteins involved in stress response (GroEL, Cpn60, RpoB, DnaK), translation elongation factor Tu, RNA polymerases and ATPases. These proteins were annotated as belonging to wide repertoire of microorganisms, most visible amongst them were *Sphingomonas wittichii, Methylosinus trichosporium*, *Bradyrhizobium, Methylocystis* sp, *Geobacter* species and members of Planctomycetales (*Planctomyces, Isosphaera*). This indicates slow revival of the microbial consortia to acetate consumption (Chourey et al. 2012).

Enrichment cultures were established using cryosol samples from 2010 and 2011 cores to determine if microbial activity could be initiated and influenced by excess amounts of different carbon sources (Tryptic Soy broth, Potato Dextrose, Glucose and R2A media). One gram of soil was suspended in 10 mL of media and the cultures were incubated at 10^{o}C for three months and then sacrificed for proteomic profiling. Remarkable microbial growth was observed in high nutrition media (TSB, Glucose, PD) whereas R2A medium, which is a minimal media selective for oligotrophic microbes exhibited modest effect in enhancing microbial activity in cryosol layers. Results suggest that AHI cryosol is very low in organic carbon required to sustain microbial activity for long time, and influx of metabolizable carbon can swiftly revive the cryosol microbial community. Names of microbes identified, varied according to database used for proteome analysis, but overall active layer included microbes involved in nitrogen cycle, few extremophiles and cyanobacteria. Permafrost layer was predominantly made up of endospore forming extremophiles along with cyanobacteria. Cyanobacteria are also known to be resistant to desiccation and starvation. Proteome characterization of different cultures led to identification of oxidative stress response proteins such as rubredoxin and rubrerythrin, cold shock and cold acclimation proteins along with acetate degradation pathway proteins hinting at anoxic conditions.

Soils were subsampled from core samples in the long-term incubation experiment. Soil samples from cores showing high CO_2 production and atmospheric CH_4 oxidation were processed for protein extraction and analyzed by MS and resulting peptides were matched against (1) a customized database containing pMMO sequences constructed from 10 metagenomes of these soils, and (2) a database containing contigs constructed from transcriptomic data of native soils collected from the study site in 2013. Although the overall cellular activity continued to be subdued in soils upon thawing, among the identified proteins were the ones related to pMMO operon of atmospheric CH_4-oxidizers. Proteome profiling of upper layers of cryosol led to identification of *pmo*B protein from the annotated pMMO operon. Identification of this protein, known for its role in CH_4 oxidation indicates microbial activity relevant to CH_4 remediation during thaw condition.

Metatranscriptomics –RNA-sequencing was performed on the surface soils collected in summer 2013 at 5-cm depth from polygon interior, polygon wedge and that under vegetation. Metatranscriptomic sequences were annotated in MG-RAST. The relative abundance of 16S rRNA genes of methanotrophs was ~100 times greater than that of methanogens (0.0545% vs. 0.00065% for polygon interior, 0.2045% vs. 0.00208% for the polygon wedge and 0.139% vs. 0.00087% for the soil beneath vegetation). Type II methanotrophs dominated the methanotrophic community. Methanomicrobia dominated the methanogenic community in the polygon wedge soil whereas Methanococci dominated in the polygon interior samples. *pmo*ABC genes are transcribed more (~183 times) in the polygon wedge samples compared to the polygon interior samples and appear to be absent in soil under vegetation. The raw transcript sequences were mapped onto a selectively assembled database of *pmoA* genes and all were closely related to Type II methanotrophs such as the atmospheric CH_4 oxidizers detected in metagenomic data and metaproteomic data. This result supports the *in situ* CH_4 uptake flux observed in the field. The lack of mcrA genes in the metatranscriptomic data suggested the low activity of methanogens in the oxic zone. Altshuler (PhD, Whyte's lab) is currently repeating the metatranscriptome analyses on triplicate 2014 samples from the same sites in a expanded study to further determine the identify active C cycling microbial communities within this permafrost site.

PLFA Analyses - Phospholipid (PLFA) analyses performed by an unsupported collaboration with McMaster University yielded the high concentrations in the permafrost. The cell concentrations inferred from the PLFA concentrations are a factor of 100x less than that of qPCR 16S rRNA copy number. If the PLFA represents the "active" microbial community then a large fraction of the DNA most likely represents an inactive microbial community that plays no role in carbon cycling. The small concentration of "active"

microorganisms could explain the lack of detection of proteins in the pristine cores and their detection only in microcosm experiments where substrate has been added.

SLIMER - Progress has been made towards developing the Scintillator-Layered Microscope for Environmental Research, SLIMER, by the Los Alamos National Lab team. Construction of the prototype was completed and imaging experiments were performed using copper masks with a 250-μm hole. The ^{241}Am α-emitter and ^{137}Cs β-emitter worked well with a 3-minute exposure. The direct imaging of a ^{14}C source with the mask has also proven successful. Further improvements on the SLIMER will be performed by Prof. Mary Kidd who has moved to join the faculty at Tennessee Technological University.

CRDS - The Princeton CRDS prototype has been used to measure the δ^{13}C and the δ^{2}H of CH_4 in atmospheric samples and a few gas samples from the thawing experiments. The current instrument at Princeton University is now capable of determining the δ^{13}C of ambient air CH_4 with a precision of ±0.8‰ and the δ^{2}H of ambient air CH_4 with a precision of ±5‰. A paper describing this result was published in Analytical Chemistry (Chen et al. 2013). A paper describing the results of CH_3D analyses is currently being prepared for submission to Analytical Chemistry.

Permafrost Flux Models - Finally as part of a Princeton University undergraduate thesis (Moch et al. 2013) a 1D model for CH_4 and CO_2 flux from active layer/permafrost has been developed and calibrated with the intact core results. This model makes use of a microbial model built upon the observed microbial community structure with depth and calibrated by the observed fluxes from the intact core experiments. Current models for microbial process, particularly for CH_4 from of thawing permafrost, were tested. These made predictions that were at odds with field observations and the intact core experiments. The new model yield results, which are compatible with flux variations as a function of temperature. A Ph.D. graduate student at Princeton University under the guidance of Prof. David Medvigy will adapt this model to EDD climate model for the Arctic.

In summary our results to date indicate that CO_2 and CH_4 fluxes during core thawing are very similar to those observed in the field. The mineral cryosols on Axel Heiberg Island are net CH_4 sinks and CO_2 emitters in contrast to organic-rich peat deposits at sub-Arctic latitudes. Twenty other 1 meter long intact cores remain at the University of Tennessee Knoxville which could be utilized for further thawing investigations if funding becomes available.

Products Delivered:

Papers:

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Abstracts:

1. Altshuler, I., Ronholm, J, Layton, A., Onstott, TC, and L. Whyte. A functionally active methanotrophic microbial community in Canadian High Arctic permafrost soils. 15th International Symposium on Microbial Ecology. (Poster Presentation) Aug. 24-29, 2014.
2. Altshuler, I., Ronholm, J., Layton, A. and L. Whyte. A functionally active methanotrophic microbial community in the Canadian High Arctic ice-wedge polygon soil. Canadian Society of Microbiologists Conference (IUMS congress), Montreal, (Poster Presentation) July 27th –Aug. 1st, 2014.
3. Ziolkowski, L.A., G. Slater, T.C. Onstott, L.G. Whyte and A. Townsend-Small. Arctic Soil Carbon Composition Governs Bioavailability. 24th Goldschmidt Geochemistry Conference, Sacramento, CA, June 8th -13th, 2014.
4. Onstott, T.C., M.C.Y. Lau, B.T. Stackhouse, D. Medvigy, Y. Chen, A. Layton, T.A. Vishnivetskaya, S.M. Pfiffner, L. Whyte, N. Mykytczuk, J. Ronholm, J. Allan, P. Bennett, K. Chourey and R.L. Hettich. An atmospheric CH_4 sink in the high Arctic and its implication for global warming. AGU Fall Meeting, San Francisco, California (poster presentation), 9th – 13th December, 2013.
5. Stackhouse, B.T., T.A. Vishnivetskaya, A. Layton, P. Bennett, N. Mykytczuk, M.C.Y. Lau, L. Whyte and T.C. Onstott. CO_2, CH_4 and DOC flux during long term thaw of high Arctic tundra. AGU Fall Meeting, San Francisco, California (oral presentation), 9th – 13th December, 2013.
6. Mykytczuck, N., B. Stackhouse, T. Vishnivetskaya, J. Allan, K. Chourey, R. Hettich, A. Layton, T.J. Phelps, S. Pfiffner, J. Ronholm, C.Y.M. Lau, A. Chauhan, D. Williams, G. S. Saaruny C., R. Sanders, S. Myneni, G. Lamarche-Gagnon, L. Ziolkowski, P. Bennett L. Whyte, and T.C. Onstott. Microbial carbon cycling in high Arctic permafrost polygons: responses to a warming climate. 5th International Conference on Polar and Alpine Microbiology. Big Sky, Montana. Invited Presentation, Early Career Researcher Award. September 8th-12th, 2013.

7. Chourey, K., M. C. Y. Lau, A. Layton, B. Stackhouse, T. Vishnivetskaya, S. Pffifner, N. Mykytczuk, L. White, R. L. Hettich, T. C. Onstott. Proteomics Reveal That Indigenous Microbial Consortia Are Revived In Thawing Permafrost And Likely Influence Carbon Cycling In The Cryosol. American Society of Microbiology (ASM), 113th General Meeting in Denver, Colorado, May 18-21, 2013.
8. Bennett, P., Stackhouse, B.T, Lamarche-Gagnon, G., Mykytczuk, N.C.S, Whyte, L.G. and Onstott, T.C. Methane and Carbon Dioxide Gas Dynamics in High-Arctic Permafrost Polygons. AGU Fall Meeting, San Francisco, California, (oral presentation), 3rd – 7th December, 2012.
9. Lau, M.C.Y., B. Stackhouse, J.M. Moch, K. Chourey, R.L. Hettich, T. Vishnivetskaya, S. Pfiffner, A. Layton, Nadia Mykytczuk, L. Whyte and T.C. Onstott. Identifying active CH_4-oxidizers in thawed Arctic permafrost by proteomics. AGU Fall Meeting, San Francisco, California, (poster presentation), 3rd – 7th December, 2012.
10. Moch, J.M., B.T. Stackhouse, M.C.Y. Lau, D. Medvigy and T.C. Onstott. Modeling CH_4 emissions from Arctic tundra: Processes behind emissions pulses and the potential for a negative feedback. AGU Fall Meeting, San Francisco, California, (poster presentation), 3rd – 7th December, 2012.
11. Sanders, R. L., Rachel L. Sleighter, T. C. Onstott, Lyle G. Whyte, Patrick G. Hatcher, and Satish C. B. Myneni. Composition of SOM in the Canadian High Arctic. 22nd Goldschmidt Geochemistry Conference, Montreal, CA, June 24-29, 2012.
12. Chourey, K., S. McGlynn, J. J. Marlow, R. Hatzenpichler, J. A. Steele, C. Pan, M. C. Y. Lau, B. Stackhouse, X. Liu, L. Whyte, G. Saarunya, A. Layton, T. Vishnivetskaya, S. Pffifner, T. C. Onstott, V. Orphan, R. L. Hettich. Proteomics Reveal Microbial Metabolic Activities Important to Environmental Carbon Cycling in Deep Sea Methane Seep Sediments and Arctic Permafrost. 112th ASM General Meeting, San Francisco, CA. Abstract #4344, (poster presentation), June 16-19, 2012.
13. Vishnivetskaya, T., A. Layton, G. Saarunya, K. Cheng, J. Murphy, S. Pfiffner, K. Chourey, R. Hettich, X. Liu, T.J. Phelps, L. Whyte, M.C.Y. Lau, B. Stackhouse and T.C. Onstott. Impact of DNA Extraction Method and PCR Amplification on Bacterial Community Composition of Arctic Permafrost and Active Layer as Defined by Tag-Encoded Pyrosequencing Analysis. 112th ASM General Meeting, San Francisco, CA. Abstract #4344, (poster presentation), June 16-19, 2012.
14. Kidd, M.F., S.R. Elliott, T.C. Onstott, S. Myeni, B. Stackhouse, S.M. Pfiffner, T. Vishnivetskaya, A. Layton, L.G. Whyte, N. Mykytczuk, J. Allan, R.C. Wilhem, R. Hettich, K. Chourey, T. J. Phelps, and P. Hatcher. Scintillator-Layered Imaging Microscope for Environmental Research. Bulletin of the American Physical Society 57: 66, 2012.
15. Lau, M.C.Y., B. Stackhouse, N.C.S. Mykytczuk, L. Whyte, and T.C. Onstott. Impact of permafrost thawing on global climate: a battle among microorganisms. International Polar Year Conference Montreal, Canada, (oral presentation) April 23-27, 2012.
16. Stackhouse, B., N.C.S. Mykytczuk, G. Lamarche-Gagnon, L. Whyte, and T.C. Onstott. Soil and Water Chemistry of Polygonal Terrain in the High Canadian Arctic International Polar Year Conference Montreal, Canada, (poster session) April 23-27, 2012.
17. Martineau, C., Pan, Y., Bodrossy, L., Yergeau, E., Whyte, L.G., Greer, C.W. Activity, diversity and community structure of aerobic methane oxidizing and carbon dioxide producing bacteria in soils from the Canadian high Arctic. High Canadian Arctic International Polar Year Conference Montreal, Canada, (poster session) April 23-27, 2012.
18. Saarunya, G.S., Vishnivetskaya T., Layton A.C., Pfiffner S.M. Development of annotated metagenome database for analyzing permafrost 'omics' data. In Abstracts of MCBIOS IX: Making Sense of the Omics Data Deluge. Annual Conference. Oxford, MS. Abstract, p. 106, (poster presentation) February 17-18, 2012.
19. Vishnivetskaya, T.A, Allan J, Cheng K., Chourey K, Hettich R.L, Layton A; Liu X, Murphy J, Mykytczuk N.C, Phelps T.J, Pfiffner S.M, Saarunya G, Stackhouse B.T, Whyte L, Onstott T.C. Microbial activity in active and upper permafrost layers in Axel Heiberg Island. AGU Fall Meeting, San Francisco, (poster presentation), 5-9 December, 2011.
20. Mykytczuk, N.C., Stackhouse, B.T., Bennett, P., Lamarche-Gagnon, G., Hettich, R.L, Phelps, T.J, Layton, A., Pfiffner, S.M., Allan, J., Vishnivetskaya, T.A., Chourey, K., Whyte, L., Onstott T.C. Gas flux dynamics in high arctic permafrost polygon and ice wedge active layer soil; microbial feedback implications. AGU Fall Meeting, San Francisco, (poster presentation) 5-9 December, 2011.

21. Saarunya, S.G., Vishnivetskaya T.A., Chourey K., Layton A.C., Pfiffner S.M. Aspects of Adaptation by Same Bacterial Genus to Diverse Environments. In Abstracts of the 3rd Annual Argonne Soil Metagenomics Workshop, Chicago, IL, (poster presentation) October 5-7, 2011.
22. Mykytczuk, N.C.S., Foote, S.J., Greer, C.W., Whyte, L.G. 2011. Subzero growth; genomic and physiological insights from *Planococcus halocryophilus* sp. nov. Or1 isolated from Canadian high Arctic permafrost. In Abstracts of the 4th International Conference on Polar and Alpine Microbiology (PAM 2011), Ljubljana, Slovenia (oral presentation), September 4-8, 2011.
23. Vishnivetskaya, T.A, Gilichinsky D.A. Biodiversity and Biogeography of Bacteria in Polar and Alpine Environments. In Abstracts of the 4th International Conference on Polar and Alpine Microbiology (PAM 2011), Ljubljana, Slovenia (oral presentation), September 4-8, 2011.
24. Vishnivetskaya, T.A., Stackhouse B., Mykytczuk N., Layton A.C., Pfiffner S.M., Phelps T.J., Whyte L., Onstott T.C. Microbial community in active and upper permafrost layers on Axel Heiberg Island. 2011, In Abstracts of 111th ASM General Meeting, New Orleans, LA, Abstract, (poster presentation) May 21-24, 2011.
25. Vishnivetskaya, T.A. Microbial communities in deep permafrost biosphere. Microperm Workshop - Circumpolar Integration of Permafrost Microbiological Studies, Potsdam, Germany. Invited plenary talk, November 8-10, 2010.

Undergraduate Theses:

1. Moch, J. - Permafrost and Global Climate Change: Novel Models and Implications for Policy and Advocacy, Senior Thesis, 2012.
2. Burton, N. – Study of gas flux rates of thawing active layer and permafrost, Senior Thesis, 2013.

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