Time-series metagenomics of experimentally warmed Alaskan tundra and Oklahoma temperate soils enables fine-resolution assessment of belowground C cycling feedbacks to climate change

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**Project goals:** The overall goal of this project is to advance systems-level predictive understanding of the feedbacks of belowground microbial communities to multiple climate change factors and their impacts on soil carbon (C) cycling processes. The specific objectives are to: (i) reveal the responses of soil microbial communities to climate warming and soil moisture alteration in both tundra and temperate grassland ecosystems; (ii) determine temperature sensitivity of recalcitrant C decomposition and characteristics of the microbial degraders; and (iii) develop integrated bioinformatics and modeling approaches to scale information across different organizational levels.

**Abstract:** Soils contain more carbon (C) in the form of soil organic matter (SOM) than both aboveground plant and atmospheric pools combined. Higher land temperatures are expected to cause the release of considerable amounts of CO₂ and CH₄ to the atmosphere, primarily through the stimulation of microbial-mediated SOM turnover. However, the direction, magnitude, and underlying basis of soil feedbacks to climate warming remain poorly understood. To this end, we have investigated microbial communities from Alaskan tundra (AK) and Oklahoma temperate grassland (OK) soils, both of which have been experimentally warmed *in-situ* (~2°C above ambient temperature) and under laboratory conditions (15 and 25°C). By combining well-replicated soil metagenomes with continuous environmental monitoring, respiration data, and soil measurements, we hope to gain an improved understanding of microbial responses to climate warming, particularly those involved in the release/sequestration of greenhouse gases.

At the AK field site, communities from deeper soil layers (45-55cm depth) were more sensitive to 5 years of field warming than surface soils (15-25cm depth) - e.g., for deep-layer communities, warming induced a significant increase to α-diversity, and community composition was largely relatable to changes in annual thaw duration, which increased by ~34 days due to warming (warming only increased surface soil thaw duration by ~8 days). Warming also increased the abundance of many SOM catabolic pathways, including those for both the labile and recalcitrant fractions of SOM. These results were also consistent with GeoChip functional gene analysis of increased ecosystem respiration reported at an earlier experimental phase (Xue et al., 2016). Sequence assembly and binning techniques allowed for the recovery of several near-complete bacterial population genomes from both AK and OK ecosystems, allowing for prediction of their metabolic lifestyles, regional prevalence, and response to elevated temperatures. Several of the recovered AK populations were regionally ubiquitous, e.g., found at several locations ~100-530 kilometers apart (Johnston et al., 2016). Consistent with the community-wide shifts mentioned above, warming favored bacterial populations encoding diverse metabolisms for recalcitrant and
labile SOM degradation, including abundant members of the community (0.25-2% of total). 5 years of similar experimental warming at the OK field site altered the functional composition of microbial communities (β-diversity distances) and also increased microbial community α-diversity. To assess shifts under more pronounced temperature changes and test for similarities between AK and OK soils, soils were incubated in the lab under 15 and 25°C. Metagenomic sequencing and assembly allowed for the recovery of several hundred-population genomes, which collectively recruited up to 75% of metagenomic reads. This allowed for more resolved associations between SOM-turnover and community composition (i.e., members responsible for these activities) to be identified. For instance, a correlation coefficient of 0.83 was obtained by relating the abundance of Acidobacteria populations to the recalcitrant fraction of SOM respired under laboratory conditions.

References

Funding statement: Our work is supported by the U.S. DOE Office of Science, Biological and Environmental Research Division (BER), Genomic Science Program, Award No. DE-SC0010715.