

**Gene- and genome-centric metagenomic analysis of complex microbial communities along a permafrost thaw gradient**

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**Project goals: As a result of climate change, large amounts of carbon sequestered in permafrost (~50% of global soil carbon) are becoming available for microbial degradation. One of the primary results of permafrost thaw is the production of the potent greenhouse gas methane, forming a positive feedback to climate change. Here, meta-omic and geochemical techniques are used to elucidate tangible links between microbiology and geochemistry along a natural permafrost thaw gradient in northern Sweden.**

We analysed ~2 Tbp of shotgun metagenomes from 214 samples taken across the permafrost thaw gradient transitioning through intact palsa, partially thawed sphagnum-dominated bog and fully thawed eriphorum-dominated fen. The samples span several depths from triplicate soil cores taken monthly during the summer thaw over several consecutive years. A comparison of metagenome-derived community profiles with 16S rRNA amplicons showed strong amplification biases against numerous lineages. The most notable differences between these profiling techniques was the absence of the candidate phylum WPS-2 in the amplicons

(despite comprising >5% of the community according to the metagenomes) and the overamplification of methanogens (up to 5-fold).

Assembly and genome binning of the metagenomes enabled the recovery of 1,529 high quality population genomes (>70% completeness & <10% contamination), including genomes from three phyla that currently lack sequenced representatives. Using a newly developed method, SingleM, which uses conserved single copy marker genes to examine community composition at high resolution, we assessed how representative the recovered population genomes were of these communities. Our results indicate that >60% of these complex microbial communities are represented by at least one population genome within the same genus.

Through the recovery of population genomes representative of the majority of microbes from these complex peat environments, we are able to couple macroscale biogeochemical measurements to predicted microbial metabolic potential. These genome-centric approaches are furthering our understanding of thawing permafrost communities, allowing us to more fully elucidate the relationships between the geochemistry, microbial ecology and global climate models.

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