***In situ* life and nucleic acid biomarker detection using the MICRO life detection platform on Mars analogue polygonal permafrost terrain in the Canadian high Arctic.**

J. Goordial[1,2], I. Altshuler[1], K. Hindosn[1], Ronan, R.[3], I. Raymond-Bouchard[1], L. Whyte[1] [1]McGill University, Macdonald campus, Ste. Anne de Bellevue, Quebec. [2]Bigelow Laboratory for Ocean Sciences, East Boothbay, Maine. [3] Oxford Nanopore Technologies, Oxford, United Kingdom

Introduction: The search for life on other solar system bodies is and will be a major focus of planetary exploration in the coming decades. The primary targets for astrobiology investigations of other solar system bodies are Mars, in the short term, and Europa and Enceladus, in the mid to longer term. Extremely cold temperatures characterize these targets, therefore the best terrestrial analogues may be the Earth's polar regions. Significant progress is being made in the development of the next generation of low cost life detection instrumentation with much smaller size, mass and energy requirements. Here, we describe use in the field of the MICRO life detection platform on ice wedge soil in polygonal permafrost terrain in the Canadian high Arctic (79ºN), a high fidelity analogue to the polygonal terrain observed on Mars[1].

The MICRO life detection platform includes (1) the CRYO-ichip for culturing microorganisms using diffusion of *in situ* nutrients on a solid media (2) a Microbial Activity Microassay (MAM) plate (BIOLOG Ecoplate) for detecting viable extant microoganisms through a colourimetric assay, and (3) the Oxford Nanopore MinION for nucleic acid detection and sequencing of environmental samples as well as the products of MAM plate and CRYO-ichip.

The small volume involved for each of these modules means that a single sample (for example a core segment or scoop of regolith) can be split for multiple analyses occurring in parallel. The ichip and MAM plate can work with environmental samples; however, analyses with the minION require a biomolecule extraction and sample preparation step prior to analysis.

While in the high Arctic, the MAM plate successfully identified an active community capable of L-serine metabolism, wells which were positive for activity were used for metagenomics sequencing with the minION to identify the active and enriched community. A metagenome on environmental ice wedge soil samples (containing ~ 10^6 cells / g) was completed while in the field, with manual biomolecule extraction prior to input into the MinION, base calling of nucleic acids was completed on a remote server, not locally, with uplink/downlink via satellite internet. Sequences from Archaea, Bacteria, Eukarya as well as viruses were detected in the Minion-generated metagenome. Validation of minION sequencing using the Illumina MiSeq platform was consistent with results obtained with the minION. After an *in situ* incubation of 1 week and 2 months in the Canadian high Arctic, we obtained ~50 microbial isolates upon return of CRYO-ichips to the laboratory. These isolates included several putatively novel strains based on the 16S rRNA gene, such as a *Pedobacter* sp. (96% closest similarity in GenBank) which we partially genome sequenced using the MinION, providing proof-of-concept that such an analysis could be carried out in the field.

This instrumentation and technology utilized in the MICRO Life detection platform is pre-existing, low cost, low mass, low volume, and offers the prospect of equipping micro-rovers and micro-penetrators with aggressive astrobiological capabilities. Since potentially habitable astrobiology targets have been identified (RSLs on Mars, near subsurface water ice on Mars, the plumes and oceans of Europa and Enceladus) [2, 3] [4], future astrobiology missions will certainly target these areas and there is a need for direct and unambiguous life detection instrumentation.

A major challenge of using such micro technologies will be developing complimentary small, robust, and automated biomolecule extraction instrumention, as well as demonstratring that such instrumentations can dectect very low levels of target molecules as expected in actual Mars, Europa, and Enceladus samples. Finally these methodologies have clear applications for 'quick and dirty' analyses of community metagenomics and assessment of microbial activity and viability in environmental field sites, including extreme sites such as the high Arctic

References:

[1] Pollard, W. et al. (2009) *Planetary and Space Science* 57 (5): p. 646-659. [2] McKay, C.P. et al., (2008) *Astrobiology* 8(5): p. 909-919. [3] Melosh, H. et al., (2004) *Icarus* 168 (2): p. 498- 502 [4] Ojha, L., et al. (2015) *Nature Geoscience* 8(11):829-32

Additional Information: This work was supported by a Canadian Space Agency Space exploration Science Definition study /CSA FAST grants, the NSERC Polar Continental Shelf program, NSERC Discovery, Northern Research Supplement, and CREATE Grants to LW, the McGill Space Institute for PDF support to JG.