Microbial characterization of groundwater from boreholes at CRL

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Purpose of study and methods

A microbiological characterization study was carried out on groundwater samples from various depths in older (CR9, CR18) and recently drilled boreholes (CRG-1, CRG-2, CRG-4A) at Chalk River Laboratories (CRL). This work was carried out as part of a technical feasibility study assessing the suitability of the CRL site to host a proposed Geologic Waste Management Facility for CRL's radioactive non-fuel waste.

A multi-analysis approach was used to characterize the water samples for microbial content. Analyses included: (1) Geochemical analysis for major cations, anions, pH, Eh and DOC. (2) Total (live + dead) and viable (live) cell counts using a number of different dyes and probes. (3) Phospholipid fatty acid (PLFA) analysis for viable cells and community structure. (4) Classic culturing for heterotrophic aerobic and anaerobic bacteria, nitrate-utilizing and -reducing bacteria and sulphate-reducing bacteria. (5) Identification of isolates using BIOLOG GEN III. (6) Identification of microbes using DNA extraction and (pyro-) sequencing.

Result and Conclusions

The water samples contained a total population of 10^4 to 10^5 cells/mL of which generally only < 1% could be cultured. However, a large percentage of the total population was viable and showed some signs of metabolic activity. Identification results for isolates showed a dominance of *Pseudomonas*, *Sphingomonas* and *Acidovorax* species. Identification based on DNA sequencing showed a dominance of different species. The combined microbial and geochemical results suggest an oligotrophic biogeochemical system in the CRL groundwater. The presence of a population of viable but not culturable cells implies that, given an increased source of electron donors (e.g., DOC) and electron acceptors (e.g., metals) leached from the waste, microbial activity could increase significantly in a potential GWMF. This could have both positive effects (e.g., lower Eh and radionuclide (RN) solubility) and negative effects (e.g., increased RN mobility, ^{14}C-containing gas production). Ultimately the biogeochemical system is expected to return to its original oligotrophic conditions but the rate at which this would occur is uncertain because waste leach rates and *in situ* microbial metabolic activity rates are unknown. This study illustrates that microbial effects need to be considered in the safety assessment of a deep geologic nuclear waste repository.

Biogeochemical Characterization of a Late Archean Sub-Seafloor Hydrothermal System, Dome Mine, Timmins, Ontario, Canada

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Much of our understanding of early life on Earth is dependant on the characterization of habitable environments preserved in Archean terrains. One such example can be found in the Tisdale mafic volcanics and hydrothermally altered metasediments of the Abitibi greenstone belt in Northern Ontario [1]. These late Archean volcanics host greenstone quartz-carbonate vein gold desposits, which are characterized by iron-carbonate alteration from low-salinity, CO_2-rich hydrothermal fluids, resulting in the precipitation of carbonates such as dolomite and ankerite.

Previous work has identified endogenous molecular fossils within the 2,770-2,685 Ma Tisdale assemblage, suggesting the presence of a subsurface hydrothermal biosphere [1]. This study is focused on a unique set of 2,690-2679 Ma crustiform banded ankerite veins within the Tisdale mafic volcanics at the Dome mine, in Timmins. This ankerite horizon provides an opportunity for the characterization of an ancient sub-seafloor hydrothermal system and its potential biosphere. We are using multiple biogeochemical techniques to characterize the system, to elucidate its environmental conditions, genesis and evolution, as well as biomarkers, and any associations with gold mineralization.

XRD and IR-spectroscopy have identified compositional variations in the carbonate speciation and minerology of the ankerite horizon. These datasets in combination with SEM and stable C- and O- isotope analysis are being used to determine the degree of hydrothermal alteration, the fluid composition and genesis, and provide environmental constraints for the system. Extracted biosignatures are being characterized by GC-MS, stable C-isotope, and ToF-SIMS analysis.

An understanding of early earth habitable environments, the developement of methods for their characterization, and the identification of biosignatures is a key aspect in furthering the search for evidence of habitable environments and extant life on Mars [2]. In particular, given the detection of Fe-Mg carbonates on the Martian surface [3,4]. As well, this research has potential implications for the development of paleobiological vectors for mineral exploration.

[1] Ventura et al. (2007) *PNAS* **104**, 14260-14265. [2] Summons et al. (2011) *Astrobiology* **11**, 157-181. [3] Ehlmann et al. (2008) *Science* **322**, 3671-1832. [4] Morris et al. (2010) *Science* **23**, 421-424.