1. **Introduction (location and background information; project history)**

Biologically active ecosystems have only recently been recognized in the Earth’s cryosphere (Priscu and Christner 2004). Potential biological communities and their role in carbon and nutrient cycles below deep permafrost have not been studied to date. This emerging view of a biologically active cryosphere and recent recognition of a potential deep cryosphere, has global repercussions on environmental fluxes and cycles considering the widespread distribution and extent of cold environments both on Earth and in other potentially habitable environments in the Solar System. Little is known regarding the microbial assemblages that comprise the cryosphere and the importance and significance of this system has only begun to be realized.

The hypersaline cold springs on Axel Heiberg Island are among the only known perennial springs flowing through thick permafrost on Earth (Andersen et al., 2002). Gypsum Hill springs located at nearly 80 N, is the only known nonvolcanic, hypersaline, sulphidic, perennial cold spring system on Earth. Drilling beneath Gypsum Hill will provide the opportunity to assess the deep cryosphere below one of the most unusual microbial communities in an extremely unique hydrogeologic setting. Such an expedition will assess how subsurface life responds to the physical and chemical extremes represented by a hypersaline, cold, anoxic brine within deep permafrost. This confluence of extreme physio-chemical parameters renders GH a truly world class geological site and presents an unprecedented opportunity to extend our knowledge of the limits of life while addressing the broader challenge of defining the distribution and extent of life on Earth.

The proposed team draws on international expertise combining multidisciplinary experts in cryomicrobiology, geomicrobiology, hydrogeology, polar geomorphology, biogeochemistry / isotopic geochemistry, and planetary science to produce a highly competent team to optimize scientific yields and capitalize on the transdisciplinary facets of the proposal and all have experience working in polar environments with many years of polar research field experience.

We propose a rigorous interdisciplinary study to assess the microbial community and geomicrobiological interface in the deep cryosphere below Gypsum Hill spring. Such a study will contribute to the global knowledge of low-biomass microbial communities and our understanding of the biogeochemical cycling of nutrients within the cryosphere. This has broad implications for understanding the distribution and significance of the as of yet poorly understood hypersaline subsurface brines in permafrost environments and may help constrain permafrost stability. Investigating the inoculation source for the GH microbial community will address the DCO decadal questions specifically “What mechanisms govern microbial evolution and dispersal in the deep biosphere?” and “What ecological rules explain deep microbial community structure?”

2. **Motivation and Goals of the Drilling Project**

The potential deep biosphere existing within and below deep permafrost is entirely unknown as is the biosphere underlying the unique cold saline springs located on Axel Heiberg Island. We propose to drill 300 to 600 m into the permafrost environment through Expedition diapir hosting the highly unique Gypsum Hill (GH) cold saline spring system in the Canadian high arctic. Samples will be collected every 25 M with higher resolution sampling through transition zones. Subsurface microbial communities and their role in C and nutrient cycles in deep subsurface permafrost cryoenvironments have not been
studied to date. GH springs (~80 N) is the only known nonvolcanic, hypersaline, sulphidic, perennial cold spring system on Earth. The springs microbial community is primarily sustained by chemolithoautotrophic primary production (sulfur-oxidization). To date, ecosystems of this type are found only in permanently dark hydrothermal vents and sulfidic groundwater but not in illuminated ecosystems. Drilling beneath Gypsum Hill will provide the opportunity to assess the deep permafrost cryosphere below one of the most unusual microbial communities in an extremely unique hydrogeologic setting. This will assess how subsurface life responds to the physical and chemical extremes represented by a hypersaline, cold, anoxic brine within deep permafrost. The major objectives of this project are to 1) determine the microbial ecology, diversity, and activity of the subsurface cryosphere; 2) develop a model for C cycling in permafrost subsurface; 3) elucidate the geomorphology/geology / thermal regime of the GH springs system subsurface environments.

This proposal to drill beneath Gypsum Hill spring represents the first study to examine the deep subsurface biology in the cryosphere. Because the presence of a cryosphere strongly affects any hydrological activity, examining characteristics of the hydrological cycle in Earth’s coldest regions may offer substantial insight not only into the widespread deep cryo-hydrogeological cycles on Earth, but also into Martian hydrology, past or present. This project has the potential to set additional limits on active microbial life on Earth and metagenomic studies comparing the surface communities with those at depth will assess "What can genomes tell us about the limits and possible origins of life?"

The highly unusual metabolic profile of the GH surface community is comparable with deep sea hydrothermal vents and subsurface microbiota. The absence phototrophy is an enigmatic feature of the GH surface metabolic profile suggesting that the microbial community may derive from the subsurface. See section 4 for more detailed information regarding microbiology. The unusual structure of the GH microbial community and paucity of studies investigating the deep cryosphere are the primary drivers of this proposal as well as better constraining the geology / geomorphology / thermal regime of this highly unique area of active salt tectonics.

Further to the enigmatic microbial community, drilling at GH presents a unique opportunity to study hydrogeologic processes in the deep permafrost and investigate a rare occurrence of anhydrite diapirs forming a well-defined wall-and-basin-structure. Axel Heiberg Island contains the second highest concentration of exposed evaporate diapirs, and is the only known location where these diapirs host perennial cold springs. The diapirs in the Expedition fjord area have been interpreted to represent a detached canopy of coalesced allochthonous evaporate sheets, see section 3 for a discussion of the regional geology. If the Expedition diapir hosting Gypsum Hill spring is indeed a second generation diapir, distinct brines flowing through evaporate deposits at distinct structural levels may give rise to spatially and temporally separated subsurface microbial communities. Drilling through the canopy diapir will yield insight into this region highly unique salt tectonics

Specific Drilling goals:
1) drill adjacent to the main GH spring outlet (GH4)
2) drill through the ~600 m thick permafrost and attempt to drill through the flank of the Expedition diapir
3) Acquire triplicate samples for biological processing and cryo-imaging from each distinct lithological or physical unit
4) Acquire detailed samples at 5-10 cm interval through transition zones
5) Use clean drilling techniques to avoid introducing contaminants into the delicate ecosystem and sub-surface aquifer system
6) Avoid contamination to preserve sample integrity

Specific Scientific goals:
1) Determine the depth of the active microbial cryosphere
2) determine differences in community structure between surface and subsurface communities
3) determine the metabolic profile of the subsurface community to see if it can help explain the anomalous profile of the surface community
4) assess the depth of the diapair structure
5) assess the existence of a allochthonous evaporate canopy and second generation diapirs
6) determine if there are separate subsurface microbial communities in pore fluid within and outside of the diapair
7) better constrain the aquifer of the GH spring system
8) explore the role of methanogenesis in the deep cryosphere to better understand the biological role of the carboniferous evaporites
9) using state of the art cryo-TEM facilities, determine the ultra-structure of pore-grain interfaces of subsurface permafrost
   a. how deep do hyersaline liquid veins exist
   b. are active cells restricted to viens
   c. how extensive are the networks
   d. how is C distributed in the fluid and around the grains comprising the permafrost.
10) constrain the thermal regime of the diapir-spring structure and potential for migration/diffusion of water and gases in shallow-deep permafrost

3. Geology/Geophysics of Study Area

Axel Heiberg Island at nearly 80°N, is located within a polar desert climate, with the surrounding landscape and conditions providing an invaluable opportunity to examine terrestrial processes in a cold, dry environment. The island is characterized by a series of forty-six diapirs of Carboniferous evaporites that have been revealed by erosion over the past tens of thousands of years (Harrison and Jackson 2014). The diapir concentration and exposure is second only to Iran and is the only such series exposed in a Polar Region. It is inferred that two structurally distinct salt-tectonic mechanisms give rise to the Axel Heiberg diapirs during the Late Triassic to Paleogene (Harrison and Jackson 2014). Evidence suggests the diapirs are still rising today. The dominant formation is comprised of first generation diapirs and associated minibasins formed during regional shortening interpreted to have detached on autochthonous Carboniferous Otto Fiord Formation (halite overlain by thick anhydrite). In the Expedition Fjord region, narrow diapirs outcrop in tight anticline cores separated by synclinal basins comprising the wall-and-basin structure. These diapirs are interpreted to represent a detached allochthonous canopy formed when
the basal diapiric roof bulged upwards, broke out and coalesced to form a canopy during Hauterivian (Harrison and Jackson 2014). Second generation diapirs rose between minibasins during burial to form the wall-and-basin structure exposed today. This region on Axel Hieburg Island is one of three known exposed evaporate canopies in the world and is the only known exposure of second generation diapirs to be exposed above the canopy (Harrison and Jackson 2014). Gypsum Hill spring is hosted in Expedition Diapir, one of the wall-and-basin structure domes sourced from the allochthonous canopy comprising a second generation diapir.

Permafrost is defined as any ground material that stays below 0°C for at least two consecutive years (van Everdingen, 2002). The depth of permafrost near Gypsum Springs is estimated to be 400–500 m based on surface temperature and regional heat flow patterns. The thickness of permafrost observed in an near by exploration well (oil and gas) was greater than 400 m, a value consistent with other exploration wells in the region that revealed permafrost depths 400–600 m (Taylor and Judge, 1976). The thin seasonally thawed active layer atop the permafrost typically measures 40–60 cm in thickness.

Permafrost exerts a profound influence on the occurrence, movement and quality of ground water in polar regions (van Everdingen, 1990). Continuous permafrost divides hydrogeologic systems into sub and suprapermafrost aquifers with permafrost actin as an impermeable barrier, the net result being that surface hydrologic and biologic systems are supplied by ephemeral waters from suprapermafrost aquifers. Deep permafrost springs can occur only where ground water is either (1) heated and reaches the surface through a hydrothermal talik, or (2) is highly mineralized, depressing the freezing point below the temperature of the permafrost to form a hydrochemical talik (Pollard et al. 1998). Cold saline groundwater springs discharge at several locations on Axel Heiberg Island (AHI) in the Canadian High Arctic and are linked to subpermafrost groundwater flow through carboniferous evaporites in areas of diapiric uplift (Pollard et al., 1999; Andersen et al., 2002). Perennial springs are extremely rare in regions of deep continuous permafrost because frozen ground restricts groundwater flow to either sub- or supra-permafrost regions, with the frozen material between serving as an aquitard and thus restricting exchange between the two systems (Williams and van Everdingen, 1973).

Described by Pollard et al. (1999), the Gypsum Hill systems comprises over 40 individual springs discharging over a band nearly 300 m long and 30 m wide at the bottom of Gypsum Hill where a sharp break in slope is coincident with an overlap of bouldery colluvial material and sandy outwash deposits. These springs flow throughout the entire year and discharge cold anoxic (mean oxido-reduction potential (ORP) of ~325 mV) brines (7.5–15.8% salts), with constant discharge temperatures ranging from ~0.5 to 6.9°C. Discharge waters are near-neutral (pH 6.9–7.5), and rich in both sulfate (2300–3724 mg l⁻¹) and sulfide (25–100 p.p.m.) (Andersen et al., 2002; Perreault et al., 2007). The main springs maintain a constant temperature throughout the year despite air temperatures that drop below -40°C during the winter. The springs are not associated with volcanic activity; the heat is provided through the local geothermal gradient. Total discharge for all of the outlets is approximately 15–20 l/s, the average outlet temperature (weighted by volume) is 2°C, and the chemistry of the solution is dominated by dissolved Na and Cl with lesser amounts of K, Ca, Mg, and SO₄ (Pollard et al., 1999). The outlets at Gypsum Hill are randomly spaced over an area of 2000–2500 m² adjacent to the Expedition River, a large glacier floodplain fed by meltwater from the White and Thompson Glaciers. Springs outlets
range from well-defined vents and pools to patches of saturated sediment. The outflow forms small gypsum and calcium carbonate coated channels and flow paths that cascade roughly 15 m to the Expedition River. Any features occurring on the Expedition flood plain are removed annually by glacial meltwater. Flow rates and temperatures are constant year round. In winter a thin veneer of snow covers some of the small higher outflows while icings and icing mounds form further downslope. A large icing (0.3–2.1 m thick) forms every year extending from the spring outlets at the base of Gypsum Hill to 700 m down stream and up to 300 m out onto the floodplain. Snowmelt and water from the active layer contributes very little to flow. Salt and gypsum precipitates cover much of the active spring area (Omelon et al., 2006a).

Although the physical and chemical characteristics of the outflows at these two sites had been well characterized, the source of the water was as of yet unknown. It has been hypothesized that the springs originate from subpermafrost salt aquifers and rise to the surface through the permafrost (4), however the source aquifer has not been defined and the subsurface hydrogeologic system is largely unknown. While Pollard et al. (1999) had originally presented five possibilities, Andersen et al. (2002) developed a combined flow and thermal model to demonstrate that the brine discharge may originate in a glacially dammed lake several kilometers away.

Down hole temperature logging that would continue promoting long term scientific benefit would prove that permafrost around the diapirs is indeed as deep or as extensive as is currently hypothesized, which would bring to light ideas about the cryosphere at this site. A drilling program will improve our understanding of the subsurface thermal regime that impacts groundwater flowpaths and microbial activity, as well as the extent of microbe-mineral interactions, rates of rock-water interactions, and carbon dynamics in a deep subsurface evaporite system. A better understanding of abiotic-biotic interface will provide the foundation to recognize and interpret potential microbial biosignatures in samples collected from cores.

There are many questions regarding the shallow and deep groundwater migration/diffusion, the C-based gases migration/diffusion and the overall structure of the thermal regime. Data on the migration/diffusion of water and gases in shallow-deep permafrost are almost completely unknown. So these are outstanding questions that can be addressed with a deep core, and it ties in well with the microbial focus. Investigating these components will generate a comprehensive understanding of microbial metabolism and the roles of these metabolites in the subsurface. For example, do microbially respired gasses stay trapped in a geological formation, or slowly diffuse to the surface? These questions can be addressed by looking at CO2-CH4 (and their isotopes) as well as a series of noble gases.

4. Previous and Relevant Work Microbiology

Relatively little is known about the diversity, abundance, and ecology of microorganisms in polar regions, where unique habitats exist. Even less is known about putative subsurface communities beneath permafrost dominated environments. To date the deepest microbial study of a permafrost environment is ~ 100 m that was performed Siberia. In addition, the NEEM ice core project in Greenland examined the surface permafrost soils underlying the Greenland ice sheet.
The microbial community and metabolic profile of surface waters from GH suggests that the community may be derived from subsurface communities inoculated into the hypersaline fluids upwellling through Expedition diapir. Alternatively, the community may be evolved from the source water reservoir, having undergone selection during the residence time of the fluids in the subsurface. The bacteria isolated from the GH springs are predominantly psychrotolerant, facultative anaerobes and grow at salt concentrations at least as high as the *in situ* salinity. *Gillisia, Psychrobacter, Marinobacter*, and *Sporosarcina* had the highest diversity of phylotypes (Perreault et al 2008). Previous works indicates that springs’ microbial community is primarily sustained by chemolithoautotrophic primary production performed by sulfur-oxidizing bacteria, even in the period of continuous illumination in the Arctic summer. To date, ecosystems of this type have been found only in permanently dark hydrothermal vents and sulfidic groundwa- ter but not in illuminated ecosystems (Perreault et al 2008).

Initial microbiological survey by Perreault et al. (2007) revealed that the majority of the 16S rRNA gene phylotypes detected within the sediment at the source of the springs were related to microorganisms involved in sulfur cycling with the major metabolic processes appearing to be the oxidation of reduced sulfur compounds. Further work identified culturable microbes from the spring sediments with autotrophic and sulfur-oxidation activities (Perreault et al., 2008), and the small amounts of hydrocarbons in gases exsolving from these springs were compositionally and isotopically consistent with microbial methanogenesis and possible methanotrophy (Perreault et al., 2007).

Analyses undertaken on grey streamers observed in run-off channels indicated that the microbial population is dominated by a genus of the Gammaproteobacteria, namely Thiomicrospira (Niederberger et al., 2008) and the biomass was proven to undertake both sulfide and thiosulfate oxidation and CO$_2$ uptake at *in situ* conditions, consistent with the presence and activity of chemolithoautotrophic sulfur-oxidizing bacteria such as Thiomicrospira. S-based chemolithotrophy has been shown to sustain microbial communities in ecosystems devoid of light such as hydrothermal vents (Jannasch and Mottl, 1985) and this non-photosynthesis-based primary production would also sustain the spring microbial communities during the months of total darkness that occur seasonally at these high latitudes. As such, the fact that these microbial structures can form and flourish under sub-zero temperatures via chemolithotrophic, phototrophic-independent means is of interest in astrobiology, particularly for the research of subsurface waters that are hypothesized to exist on Mars (Pollard et al 2009).

In addition to sulphur oxidation, two lines of evidence suggest that methanogenesis may occur in the Expedition Fjord spring sediments (Perreault et al 2007). First, low concentrations of methane in the spring waters at Gypsum Hill has been detected and second, the most abundant *Euryarchaeota* phylotype from both spring libraries was associated with the *Methanosarcinales* cluster in the phylogenetic trees, and one phylotype was 99% identical to psychrotolerant methanogens (*M. burtonii* and *M. alaskense*) that can use methylamines for growth (Franzmann et al 1992, Singh et al 2005). As sulfate reducers outcompete methanogens for most energy sources, the persistence of methanogens in saline environments where sulfate is not limiting is associated with the utilization of noncompetitive substrates such as methylamines (Perreault et al 2007). The predominance of CH$_4$ in the springs rather than a CH$_4$– higher hydrocarbon mixture and enriched $\delta^{13}$C values suggests a microbial origin for these gases (Perreault et al 2008). The significant
variation in $^{13}$C$_{CH_4}$ values observed over time indicates, however, that the system is more complex, with methanotrophic activity as the most likely cause of the enriched $^{13}$C values (Whiticar 1999). The highly reduced environment in the springs (-283 to -324 mV) should allow methanogenesis, and 16S rRNA gene sequences related to *Methanosarcinales* have been detected (Perreault et al 2007). Methanotrophy in the GH springs may be performed by aerobic methanotrophs but could also be the result of anaerobic methane oxidation, which is catalyzed by *Archaea* phylogenetically related to *Methanosarcinales* in association with deltaproteobacterial SRB (Boetius et al 2000, Orphan et al 2001). Isolates or 16S rRNA gene sequences directly related to known aerobic methane-oxidizing bacteria or anaerobic methane-oxidizing archaea have not yet been detected in the GH springs, however, a high proportion of sequences related to *Methanosarcinales* and deltaproteobacterial sulfate reducers have been identified (Parraduel et al 2008). Detailed microbial studies will assess the diversity and distribution of the communities beneath the spring and shed light on the unique metabolic profile of the spring. Integration with geochemical and hydrogeology studies will allow elucidation of biogeochemical processes within the deep cryosphere with implications for carbon cycling. Detailed down hole isotopic profiles will help unravel the connections between abiotic carbon sourced from the expansive carboniferous evaporites and biologically mediated carbon pathways addressing the role life plays in shaping carbon transformations in the subsurface cryosphere. Investigations into microbial carbon cycling in deep carboniferous evaporite permafrost has not previously been studied and has the potential to significantly enhance our understanding of how deep life and its influence on the carbon cycle interact with the surface world in periglacial environments. There is isotopic evidence that FeS coming from the spring outlets is biogenic; many groundwater systems follow a redox tower however whether FeS precipitation occurs in sediments near the outflows or at depth has not been answered and can be addressed through the proposed drilling.

Similar to aphotic ecosystems such as deep-sea hydrothermal vents and sulfidic groundwater, chemolithotrophs may be the main primary producers in the GH springs’ ecosystem. There are several lines of evidence supporting this hypothesis as follows: (i) an abundance of chemolithoautotrophic sulfur oxidizers were found in both this culture-dependent study and the initial culture-independent study (Perreault et al 2007); (ii) photoautotrophic phylotypes of sulfur oxidizers or cyanobacteria were not detected in the culture-independent study despite the high sulfide concentration and the continuous illumination during the sampling period; (iii) eukaryotic DNA was not detected in the GH spring sediments by using diverse sets of 18S rRNA gene primers, and phototrophic eukaryotic cells were not seen by microscopic observation indicating the absence of phototrophic eukaryotes; (iv) chlorophyll was not detected over the surface of the carbonates from ~100 spring locations by using a pulse amplitude modulation fluorometer (Anderson, 2004); and (v) the only possible photosynthetic organisms detected were the heterotrophic bacterial isolates possessing a *pfuM* sequence. However, such photoheterotrophs do not use carbon dioxide as their sole carbon source: they use organic compounds from the environment for their carbon requirements. Thus, unlike photoautotrophs, they do not provide net supplies of carbon to the ecosystem (Kepkay et al 1979) and may represent wind-borne environmental contamination.

Endolithic microorganisms (including bacteria, eukarya, and archa) occupy a unique niche in terrestrial habitats by living beneath rock surfaces as interstitial habitants of
cracks, fissures, and pore spaces between mineral grains (Goublic et al., 1981). Geomicrobiology as they are found at the organic–inorganic interface and have profound implications for our understanding of dissolution and precipitation reactions that lead to both the destruction of lithic substrates as well as the creation of authigenic minerals to produce biosignatures. Endolithic microbial communities in cold desert environments define the limits of life in polar regions, and have repeatedly been suggested to be one of the most likely places for viable life on Mars as it entered its final cooling period (Friedmann and Koriem, 1989; Friedmann, 1982; Mackay, 1999). A detailed investigation of endolithic cryoenvironments will add significant to our understanding of how life influences transitions between abiotic and biotic realms.

The ultimate astrobiology goal pertinent to drilling GH is to evaluate the potential for the occurrence of life and the preservation of different types of fossil biosignatures throughout Martian history (Fairén et al., 2005). Terrestrial analogues, such as Gypsum Hill spring are characterized by extreme conditions that are challenging for terrestrial life, e.g. temperature and salinity. By life processes operating under extremophilic conditions, we gain valuable information with respect to the limits of life on Earth, as well as the capability of organisms to adapt and survive in extreme environments. Obtaining detailed knowledge of the biological, geochemical, and geological processes in selected Mars analogues, such as Gympsum Hill will allow us to better interpret past, present, and future Mars mission results and improve the design and execution of future orbiter and lander missions. In addition, the study of terrestrial analog sites, especially sites with permanent ice, has relevance to planetary protection issues (MEPAG SR-SAG, 2006; Fairén et al., 2010).

5. Global Importance of Study Area

As discussed in sections three and four, Gypsum Hill spring is a highly unique biological and geological system. It is one of the few perennial cold springs in the world, the only known such spring to harbor a microbial community dominated by chemotrophic metabolism and is set in the only above canopy exposure of second generation evaporate diapirs in the world. Drilling at GH will provide the first opportunity to investigate the microbial communities of deep cryosphere in addition to shedding insight into a unique system of active salt-tectonics. Logistically, GH is located in close proximity to the McGill Arctic Research Station (MARS) within 40 min walking distance and 10 to 15 min by snow mobile. MARS is located 8 km inland at Expedition Fjord, Nunavut, on Central Axel Heiberg Island in the Canadian High Arctic (approximately 79º26’N, 90º46’W). Established in 1960, MARS is one of the longest-operating seasonal field research facilities in the high Arctic. The station consists of a small research hut, a cook house, and two temporary structures. MARS can comfortably accommodate up to twelve persons. MARS provides access to glacier, ice cap, and polar desert environments. The surroundings are mountainous and glaciated. MARS has the longest continuous mass balance record for any high Arctic glacier (White Glacier). Some of the most detailed environmental information in the Arctic, including topographic map data, have been collected at this station. Current research activities include glaciology, climate change, permafrost hydrology, geology, geomorphology, limnology, planetary analogues, and microbiology. Mars has hosted numerous international collaborations through the activites of Whyte, Pollard, Omelon, and Andersen.
In addition to the terrestrial insights offered by the proposed research, Gypsum Hill Spring and the Axel Heiburg environment is a world-class Mars analogue site offering unprecedented astrobiological opportunities. If extant life exists on Mars, it is likely in the subsurface, taking refuge from harsh surface conditions. Liquid water on Mars today is likely in the form of subsurface eutectic brines in a spatially restricted hydrogeological cycle existing in thick permafrost. The hydrogeology of Axel Heiburg Island represents a terrestrial analogue approximating the putative physical and geochemical conditions existing on during the transition from a cold and wet Mars during the Noachian to the hyperarid conditions that characterize Mars today (Fairén et al 2010). During this time the Martian hydrosphere shifted towards a global cryosphere and surface habitability conditions deteriorated pushing putative life towards subsurface niches (Fairén et al 2010). Permafrost dominated paraglacial environments therefore serve as analogue research sites for this phase of Martian habitability, the combination of liquid brines and deep permafrost have earned Gypsum Hill spring a world-class reputation as a Martian analogue (e.g. Fairén et al 2010). To date, the extent of the biosphere in permafrost environments is unexplored and critical to assessing the potential for both extant Martian life and the preservation of biosignatures in such environments. Another significant unknown concerns the mechanisms by which microorganisms and biosignatures survive for long periods of time in ice. There is emerging evidence that, even at low temperatures, organisms trapped in ice are capable of metabolic activity that could be used to counteract racemization or repair DNA and other structures (Carpenter et al., 2000; Price and Sowers, 2004; Tung et al., 2005, 2006; Rohde and Price, 2007), but more investigation is warranted (Fairén et al 2010).


The drill site selection and proposed work indicated in Section 2. “Motivation and Goals of the Drilling Project” and is aligned with our primary goals to determine the depth of permafrost and therefore the extent of microbial activity. Tightly coupled to this would be a determination of the geochemical environment supporting life in the deep subsurface, which is controlled by rock-water and microbe-mineral interactions occurring both within and beneath permafrost.

The microbial ecology, biodiversity and activity will be assessed in selected cores by culture dependent and culture independent methodologies by Whyte, Parro, and Onstott using methods already established in their labs. Whyte will determine microbial biomass in the samples by direct microscope counts; microbial activity will be determined through microcosm assays using radioisotope labelled tracers as previously performed; microbial community composition and function will be assessed through parallel metagenomic and metatranscriptomic analyses of the cores samples as well as 16S rDNA community profiling. Whyte will also attempt to isolate and characterise microorganisms (heterotrophic, chemolithotrophic) using his cryomicrobiology facilities at McGill University. Parro will also examine in situ microbial diversity and biomarkers with LDChip and SOLID instrument; ii) Determining main anions by Ion Chromatography (IC, in the lab). In a single IC run we can detect: acetate, formate, propionate, oxalate, Cl-, F-, nitrate, nitrite, phosphate, sulfate (run every 5 m (or more or less) can be done to have a whole profile along the borehole); and iii) Biochemical (proteins, sugars, enzymatic activities) and
organic extraction (lipids) and analysis by current biochemical techniques and GC/MS, HPLC.

7. Expected Benefits of the Proposed Work

A rigorous drilling expedition is fundamentally necessary to access the subsurface cryosphere. Outstanding globally relevant scientific questions such as the depth of the active microbial community and thermal regime in permafrost will be answered. Drilling in permafrost presents unique infrastructural challenges. Development of this project has societal implications for northern communities and projects with opportunity for industry collaboration. The requirement for northern drilling are challenges shared by both the mining and scientific communities alike. Development of clean drilling huge implications for life detection missions. The investigation of subsurface life in permafrost areas has direct relevance to the search for life on other worlds in our Solar System. The worlds of most interest: Mars, Europa and Enceladus are all cold and in each case it is clear that the search for life, or evidence of past life, is best conducted by investigating the subsurface environments. ExoMars, scheduled to launch 2018, is the first mission to explore the Martian subsurface, we will exploit the timely synergy between analogue environments. Drilling below GH will explore the active enigmatic processes that give rise to one of the most geologically and biologically unique spring systems on Earth. The very nature of this proposed project is aligned with the Deep Life mandate to “assess the nature and extent of the deep microbial and viral biosphere.” Not only will a drilling campaign at GH offer enormous insight into the limits of life and/or preservation of biological mechanisms on Earth, but it will also serve as technological development for current and planned life detection missions to Mars. The extreme nature of the environment and links to on-going planetary missions are of great public interest. Live uplinks, public lectures, Twitter feeds, and Google Earth uploads will be explored as modes for public outreach and education.

8. Project Management (including PIs and their roles and responsibilities)

Whyte will be the Principal Investigator of the project and logistical and science teams for the project will be formed to develop and manage the drilling and scientific objectives of the project.

9. Project Collaborators/ Science Team

Lyle Whyte, McGill University – cryomicrobiology, environmental microbiology, astrobiology.

Tullis Onstott, Princeton University – subsurface geomicrobiologist, geochemist.

Wayne Pollard, McGill University – polar geomorphology, permafrost geomorphology.

Chris Mckay, NASA Ames – astrobiologist, planetary scientist, planetary geology.

Chris Omelon, U. Texas, - microbe mineral interactions, aqueous geochemistry of polar environments.
Dale Anderson, SETI, USA – polar geomorphologist, isotopic geochemistry

Victor Parro, Spanish Astrobiology Center – microbial ecology, subsurface microbiology, astrobiology.

Denis Lacelle, U. Ottawa – polar geophysics, geomorphology, periglacial geomorphology, environmental geochemistry

Haley Sapers – geomicrobiologists, microbial and geochemical microscopy

Marcos Zentilli, Dalhousie U. – geology, isotope geochronology

10. Time Table

2015 – submit final proposal, if selected, apply for appropriate permits licenses, apply for PCSP logistical support for a 2016 drill trip to AHI, science time meeting to finalize 2016 field trip to AHI; testing trip at the drill site using Whyte’s portable permafrost drill (max.

2016 April – Primary Drilling expedition to the AHI during late winter conditions., initial on site core logging, in situ observations and measurements. Installation of down hole sensors for continuous measurements of physical / chemical measurements; Transport and curation of cores back to McGill University, storage of cores in the cryochambers of Whyte and Pollard. Post –Drilling scientific meeting to prioritize sample distribution and subsequent analyses.

2016 – 2018 – microbial, geochemical/isotopic analyses of core samples. Annual progress meetings to address the progress of the project and assess the overall objectives; report results at scientific meetings and peer reviewed publications in the domains of microbial ecology, geomicrobiology, global C flux and global warming, periglacial geomorphology, geology, planetary exploration /astrobiology.

Project Costs – will be very high, and logistically challenging as are all research projects in the high arctic. Field project costs will be leveraged through PCSP support (Whyte, Pollard); it is also anticipated that the science costs will be mostly covered by the applicants current grants and resources and that this project has significant potential to receive complementary funding from the Canadian Space Agency and NASA because of its relevance to planetary exploration, especially the proposed drilling activities that will be part of the ESA ExoMars and NASA 2020 mission.

11. References

TBD

12. Appendices

TBD