Glacial secrets uncovered: Revealing the modes of survival of metabolically active microbial communities entrapped in polar glacial ice

Brady RW O'Connor‡, Lyle G Whyte‡

‡ McGill University, Montreal, Canada

Corresponding author: Brady RW O'Connor (brady.oconnor@mail.mcgill.ca)

Abstract

Glaciers, once dismissed as inhospitable environments, have been overlooked in scientific investigations. Previous studies have primarily focused on the supraglacial (cryoconite holes, snow, and meltwater) and subglacial (bedrock and soils, among others) environments, neglecting the englacial (inside ice) realm. Despite evidence demonstrating the survival of cells in glacial/sea ice (Christner 2000, Junge et al. 2002, Miteva et al. 2004, Miteva and Brenchley 2005) and theoretical predictions and indirect evidence hinting at active microbial communities within glacial ice (Krembs et al. 2002, Junge et al. 2004, Price and Sowers 2004, Tung et al. 2005, Tung et al. 2006, Rohde et al. 2008), the englacial environment has remained largely unexplored.

Recognizing that englacial ice hosts potentially active microbial communities carries significant implications for the future of these habitats in the face of escalating global warming and glacial retreat. As glaciers rapidly melt due to the effects of global warming, the liberation of these microbial communities will undoubtedly exert profound effects on local ecosystems and biogeochemical cycles, presenting an array of unknown consequences. Furthermore, considering the ability of microbial communities to persist in such extreme conditions on Earth, they become intriguing subjects for the search for life on celestial bodies such as Mars, Europa, Enceladus, and Titan, all of which house vast ice deposits.

However, several fundamental questions persist. The extent of metabolic activity in glacial ice remains uncertain, as does the identification of microorganisms capable of sustaining metabolic processes. Most importantly, understanding the survival strategies employed by these organisms in such an extreme environment remains unknown.

To answer these questions, we present metagenomes and what we believe to be the first metatranscriptomes ever analyzed from glacial ice. We have developed a method which allows us to melt ice cores without altering the mRNA profile of the microorganisms within, allowing us to directly determine how microorganisms are able to survive in such

a hostile environment. One-to-two-meter cores were taken from the surface of White Glacier, Axel Heiberg Island and from the Devon Island ice cap, both in the Canadian High Arctic. A depth of 70 – 90 cm and 131 – 151 cm was chosen for analysis from White Glacier and Devon Island respectively. In the lab, to remove surface contamination from the cores, the outer 0.5 cm of the cores was removed, and the inner cores were sprayed with 70% ethanol. The decontaminated core subsections were melted at 4°C directly into DNA/RNA Shield (1:1 ratio) which preserved the microbial communities on contact, preventing changes to the metagenome or metatranscriptome during melt. Melted samples were filtered and nucleic acids extracted before DNA and RNA sequenced on an Illumina NovaSeq 6000 sequencer. Sequencing yield from Devon ice cap was low, resulting in few metagenomic and metatranscriptomic sequences however White Glacier produced a metagenome of 46 million reads and a metatranscriptome of 56 million reads. These data revealed that White Glacier is dominated by Cyanobacteria and Actinobacteria and the Devon ice cap is dominated by Proteobacteria. Furthermore, metatranscriptomic analysis of microorganisms from White Glacier revealed a metabolically active microbial community reliant on oxygenic photosynthesis, and carbon fixation via the Calvin and 3-hydroxypropionate Cycles. Transcripts related to aerobic respiration, aerobic carbon monoxide oxidation, sulfur oxidation, nitrite oxidation, nitric and nitrous oxide reduction and anoxygenic photosynthesis were also present. Cold adapted microorganisms possess many mechanisms to deal with low temperatures and the microbial community of White Glacier is no different. Cold response genes were highly expressed, principally membrane and peptidoglycan modifying proteins which increase membrane and cell wall fluidity at low temperatures, translation and transcription factors which increase the efficiency of protein synthesis at low temperatures, and cold shock proteins which stabilize RNA at low temperatures.

Two high and five medium quality metagenome assembled genomes (MAGs) were also recovered from the White Glacier ice core, including a 99.86% complete Coleofasciculaceae Cyanobacterial genome with transcripts mapping to its genome related to aerobic respiration, oxygenic photosynthesis, carbon fixation (Calvin cycle) and nitrous oxide reduction. Transcripts related to cold response included those from categories such as cold shock proteins, DNA repair, membrane and peptidoglycan alteration, osmotic stress, and transcription and translation factors. Transcripts involved in DNA replication even mapped to the genome of this organism indicating it was capable of cell division in the ice.

Overall, our research suggests that englacial ice can support an active microbial community where Cyanobacteria act as primary producers, generating energy from photosynthesis and fixing carbon from trapped atmospheric CO_2. Carbon monoxide is also used as an energy source. As expected, cold adaptation genes are widespread and highly expressed indicating a community highly adapted to life in glacial environments and which may even be capable of growth. This research is significant because it presents the first metatranscriptomic profile of microorganisms trapped within glaciers, challenging preconceived notions about the habitability of ice. These results

carry profound implications for the field of astrobiology and the quest to uncover signs of life within Martian glaciers or the ice-covered surfaces of Europa, Enceladus, and Titan.

Keywords

Cryobiology, Astrobiology, Microbial Ecology, Ice, Glaciers, Arctic

Presenting author

Brady RW O'Connor

Presented at

Oral Presentation, I am open to poster presentation. My abstract fits best in Part 3: Astrobiology and Earth Analog Settings in the theme area "Icy Environments".

Hosting institution

McGill University

Author contributions

Brady RW O'Connor contributed to the abstract by collecting samples in the field, performing all laboratory work, analyzing data and writing the abstract. Lyle G Whyte contributed to the abstract by providing supervision, obtaining funding and helping collect samples in the field.

Conflicts of interest

The authors have declared that no competing interests exist.

References

- Christner B (2000) Recovery and Identification of Viable Bacteria Immured in Glacial Ice. Icarus 144 (2): 479-485. https://doi.org/10.1006/icar.1999.6288
- Junge K, Imhoff F, Staley T, Deming W (2002) Phylogenetic Diversity of Numerically Important Arctic Sea-Ice Bacteria Cultured at Subzero Temperature. Microbial Ecology 43 (3): 315-328. https://doi.org/10.1007/s00248-001-1026-4
- Junge K, Eicken H, Deming J (2004) Bacterial Activity at −2 to −20°C in Arctic Wintertime Sea Ice. Applied and Environmental Microbiology 70 (1): 550-557. https://doi.org/10.1128/aem.70.1.550-557.2004

- Krembs C, Eicken H, Junge K, Deming JW (2002) High concentrations of exopolymeric substances in Arctic winter sea ice: implications for the polar ocean carbon cycle and cryoprotection of diatoms. Deep Sea Research Part I: Oceanographic Research Papers 49 (12): 2163-2181. https://doi.org/10.1016/s0967-0637(02)00122-x
- Miteva V, Brenchley J (2005) Detection and Isolation of Ultrasmall Microorganisms from a 120,000-Year-Old Greenland Glacier Ice Core. Applied and Environmental Microbiology 71 (12): 7806-7818. https://doi.org/10.1128/aem.71.12.7806-7818.2005
- Miteva VI, Sheridan PP, Brenchley JE (2004) Phylogenetic and Physiological Diversity of Microorganisms Isolated from a Deep Greenland Glacier Ice Core. Applied and Environmental Microbiology 70 (1): 202-213. https://doi.org/10.1128/aem.70.1.202-213.2004
- Price PB, Sowers T (2004) Temperature dependence of metabolic rates for microbial growth, maintenance, and survival. Proceedings of the National Academy of Sciences 101 (13): 4631-4636. https://doi.org/10.1073/pnas.0400522101
- Rohde R, Price PB, Bay R, Bramall N (2008) *In situ* microbial metabolism as a cause of gas anomalies in ice. Proceedings of the National Academy of Sciences 105 (25): 8667-8672. https://doi.org/10.1073/pnas.0803763105
- Tung HC, Bramall NE, Price PB (2005) Microbial origin of excess methane in glacial ice and implications for life on Mars. Proceedings of the National Academy of Sciences 102 (51): 18292-18296. https://doi.org/10.1073/pnas.0507601102
- Tung HC, Price PB, Bramall NE, Vrdoljak G (2006) Microorganisms Metabolizing on Clay Grains in 3-Km-Deep Greenland Basal Ice. Astrobiology 6 (1): 69-86. https://doi.org/10.1089/ast.2006.6.69