

WESTMINSTER COLLEGE

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Abstract

Gypsum (CaSO₄ \cdot 2H₂O) and mirabilite (Na₂SO₄ \cdot 10H₂O) are common precipitates at the Great Salt Lake (GSL). NASA's Mars Exploration Rover, Opportunity, found veins of gypsum deposited by water in 2011 and gypsum has been detected on Mars as early as 2005 by the ESA's Mars Express Orbiter. On Earth, gypsum is formed in hypersaline environments, in minerals left behind when water evaporates, trapping microorganisms in fluid inclusions. Likewise, GSL mirabilite develops when mineral-rich groundwater meets cold winter air to create crystalline structures. While there is no physical evidence of mirabilite on Mars, HiRISE Image data show white mounds that some experts hypothesize are related to saline ground waters. Gypsum obtained from Great Salt Lake was used to develop a method to extract halophilic archaea and culture it in the lab. The method obtained could be used to isolate potential microorganisms present in gypsum samples from Mars. Mirabilite is currently under investigation for its ability to enrich halophilic life.

Background

NASA's Rover Opportunity discovered gypsum mineral deposits in 2011 at Meridiana Planum on Mars. More recently, mound-like structures in the mid to high latitudes of Mars are hypothesized to emerge from subsurface fluid eruptions^{1,2}. These geomorphic structures and mineral deposits are suggestive of an ancient saline lake that is now evaporated. ^{3,4} Halite and gypsum are known to create fluid inclusions during their formation which can trap and store biological materials, providing shelter from physical and chemical damage. ^{5, 6, 7} Microscopy has suggested the presence of halophilic microbiota living in the saline spring water that feeds the mirabilite mounds. Mirabilite is currently under investigation for its ability to enrich halophilic life.

Great Salt Lake may serve as an analog to the evaporated salt lake regions of Mars. GSL has no outlets and has large salt flats which are halite deposits formed by the evaporation of the ancient lake Bonneville.⁸ Several studies indicate that halophiles can survive over geologic time when preserved in halite crystals.⁹ Therefore, not only is the study of GSL mineral evaporites important to understanding the evaporites on Mars, but the also the study of the entrapped halophiles will provide a model for the preservation of extant microorganisms trapped in Mars mineral evaporites that are resistant to desiccation, UV damage, and extreme osmotic conditions.

Materials & Methods

- Media: 23% Minimal Growth Media (MGM).⁸
- DNA extraction was done using the FastDNA Spin Kit for Soil.⁹
- Taq PCR kit.¹⁰
- Primers 1HK (5' ATTCCGGTTGATCCTGCCGG 3') and H589R (5' AGCTACGGTTTAGGC 3').^{11, 12, 13, 14}

Various cultivation methods were performed for gypsum crystals comparing the effectiveness of halophilic archaea growth for non surface-sterilized gypsum and surface sterilized crystals with 100% ethanol. The methods tested included placing crystals directly into media; crushing the crystals with a mortar and pestle then placing in broth; rubbing gypsum crystals directly onto agar plates; sprinkling crushed crystals directly onto plates; and finally dissolving gypsum in water based on its solubility value, then combining with broth in dilutions of 1:4, 1:2, and 1:1. Incubation temperature was 37°C.

The DNA from colonies was isolated. PCR amplification was done for the archaeal 16SrRNA gene using protocol.^{16, 17} The PCR product was amplified and observed on a 1.2% agarose gel. The products were further cleaned with a QIAquick PCR Purification Kit and submitted to the Center for Integrated BioSystems at Utah State University for sequencing.¹⁷

Mirabilite sampling took place on the west shore of Antelope Island from mineralrich spring water and resultant crystals (Fig. 1). The samples were subjected to microscopy and microorganisms were observed consistent with the sizes of both eukaryotic algae (e.g. Dunaliella sp.) (Fig. 2), bacteria, and archaea.

Mineralogy of Great Salt Lake: An Analogue for Martian Evaporites

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Results



Figure 1: Drilling into mirabilite mound resulting in a flow from underground saline spring water (top). Green mirabilite pool (bottom).



Figure 4: Neither the crushed samples placed in broth or sprinkled on plates showed any halophilic growth. The crushed samples dissolved in water and placed in broth did not show any growth either.

Figure 3. Gypsum crystal collected from Great Salt Lake North Arm near the artwork, Spiral Jetty (Robert Smithson, 1970). Samples placed directly into broth became turbid and light pink after two weeks. This was the only cultivation method that resulted in halophilic growth



Figure 5: Plating the turbid samples of gypsum crystals from the aqueous MGM resulted in growth of bright red colonies after incubation for about 3 weeks.



Figure 2: Microscopy from mirabilite samples shows eukaryotic algae (top right). Horned mirabilite crystal (bottom right)





Figure 6: Gel amplification of the PCR product indicated that the sample in lane 2 contained a roughly 550 bp product (indicated). Lane 1 contains a 100 bp Ladder (NEB). The sample was sent in for sequencing. Results were input into NCBI BLAST which resulted in a 99% match with Halobacterium noricense strain DAB-31 isolated from Black Lake in China using 16s rRNA.¹⁹





Our studies indicate the most successful method for cultivation from gypsum was placement of a surface sterilized crystal into broth. This method resulted in the growth of microorganisms that could not be identified using the halophilic archaea primers (perhaps these are bacterial isolates). Additionally, microscopy indicated the presence of microbiota in saline mirabilite spring water. These have not yet been identified as bacteria or archaea and require cultivation, isolation, and amplification. Further experiments will be done to improve halophilic archaea cultivation from gypsum, which will include methods such as isolation and culture from fluid inclusions in gypsum crystals. The successful method of halophilic archaea isolation can be used for analysis of gypsum or mirabilite returned from Mars, following the Mars 2020 Mission, to potentially cultivate any microorganisms present. The work presented here is being published as a part of a larger manuscript. ²⁰

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Figure 7: Comparison of gypsum deposits on Great Salt Lake (right) and Mars gypsum deposits imaged by Curiosity (left). Photo Credit: Scott Perl

Figure 8: Comparison of hypothesized Martian mirabilite imaged by (left) and GSL mound (right).² Photo Credit: Calli Cahill

Conclusions

Acknowledgments

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