

A Survey of Halophilic Microorganisms at the Mars Desert Research Station

John Thaler

Abstract

A variety of observational techniques will be used to determine the halophilic microorganism diversity and abundance in the MDRS region. The field component of this research will be conducted during the two week Expedition Beta mission. This will involve sampling missions to collect soil and mineral samples from areas identified as potential hypersaline habitat. The collection and analysis of soil and mineral samples will inevitably be a part of future manned missions to Mars. Hypersaline environments surrounding MDRS will be characterized using measurements of pH, salinity, temperature and moisture content (soil samples). Initial analysis of biological samples will involve separation and purification of colonies from the raw sample. Cultured samples will be shipped offsite for additional isolation, purification and identification. Aerial photographs of the sites taken by the Aerial Recon Vehicle 'Red Flyer' will be compared for halophilic microorganism density and diversity to determine if aerial identification is feasible. This project is a component of a larger initiative, the Mars Analog Microbial Observatory.

LITERATURE REVIEW

Halophilic microorganisms are defined as organisms that have optimal growth in concentrations above 0.2 M NaCl and some are known to thrive above 5 M NaCl.¹ These extremophiles have been found in a variety of microenvironments including salt lakes and brines, saline soils, cold saline environments, alkaline saline habitats, and in salted fish, meat and other foods.² Originally it was thought that Archaea dominated the hypersaline environments but it is now known that even in extremely saline conditions near the saturation point of NaCl there are examples of halophiles from a wide variety of taxa including eukaryotes (rotifers, tubellarian worms, copepods)¹, protozoa (*Porodon utahensis* and *Fabrea salina*)¹, fungi (*Cladosporium glycolicum*)³, cyanobacteria (*Aphanothece halophytica*)³, Archaea (*Haloanaerobacter chitinovorans*)¹, Bacteria (*Ectothiorhodospira halochloris*, *Salinobacter* sp.)^{3,4}, diatoms (*Amphora coffeaeformis*)³, and green algae (*Dunaliella salina*)³. Identification and classification of halophilic microorganisms is generally achieved through comparison of the 16S rDNA sequence⁵ or is based on chemotaxonomic criteria.⁶

Halophiles employ a wide variety of mechanisms to survive in the hypersaline environment. The most common methods of avoiding desiccation involve using organic osmotic solutes such as glycerol, glycine betaine, or amino acids to prevent water loss.² Halobacteria also have a light-driven chloride pump called halorhodopsin that helps maintain the cell's ion concentrations.¹ The halophilic flora and fauna of the two largest hypersaline lakes, the Dead Sea in the Middle East and the Great Salt Lake in Utah, have been the most studied but the halophiles of small evaporation ponds have also been examined and characterized.¹ At the Utah Mars Desert Research Station (MDRS) several hypersaline environments have been identified by previous MDRS crews, specifically the 5-day Everest Rover Mission that identified Salt Wash as a potential halophile sample site.⁷

It has been proposed that any near-surface water on Mars would be a highly concentrated brine solution due to the combination of the low temperatures, salty soils and ambient pressure at low elevations on Mars.⁸ This suggests that hypothetical microorganisms existing on Mars could be similar to terrestrial halophiles and techniques developed to identify halophiles on Earth could be used to find similar microorganisms on Mars.⁸

PROBLEM STATEMENT

To date there has been no systematic survey of halophilic microorganisms in environments surrounding the MDRS for the Mars Analog Microbial Observatory Project.^{9,10} Samples need to be collected from identified hypersaline habitats and the halophilic microorganisms and their environments need to be characterized. Furthermore, comparison with aerial observations could provide a remote method of detecting hypersaline habitats.

REQUIRED RESOURCES

1. Physical and topographic maps* of the area surrounding the MDRS, including the locations of suspected hypersaline environments
2. Site Sampling Equipment: digital camera*, GPS*, pH meter[†], digital thermometer[†], conductivity meter[†], analytical balance*
3. Halophile Culture Equipment: culture plates*, test tubes*, culture medium^{11,*}, autoclave[†], 37°C incubator*, microscope*

* denotes equipment available at MDRS

★ denotes applicant's personal equipment

† denotes equipment available on loan from Professor Shannon Rupert Robles at San Diego Miramar College, San Diego.

METHODOLOGY

1. Suspected hypersaline habitats will be sampled and environmental conditions measured (pH¹², salinity¹³, temperature, and soil moisture content¹⁴).
2. Multiple soil and mineral samples will be transported back to the habitat for further analysis and stored in sample containers.
3. Samples will be stored at the local ambient temperature at an outdoor storage location until they are selected to be cultured.
4. Selected samples from each site will be cultured on complex media¹¹ at 37°C.
5. Viable colonies will be isolated and observed with a microscope using basic staining techniques to obtain a preliminary identification.
6. At the end of the expedition the cultures and unprocessed samples will be shipped to Vuong Nguyen, at the Scripps Research Institute in La Jolla, for further analysis and identification. The samples will be cultured in a series of media with increasing concentration of NaCl, from 1M to 5M to determine the range of halophilic organisms present.
7. Species location and environmental conditions will be correlated with aerial and satellite photographs.

PROPOSED ANALYSES

Analysis will consist of systematically identifying halophilic microorganisms and the associated environmental conditions at sites around MDRS for inclusion in the Mars Analog Microbial Observatory Project. The comparison of aerial photographs with habitat and species present to determine if halophilic populations can be detected remotely is also of interest.

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