
Introduction: The search for extant life in our solar system includes the search for molecules specifically associated with biology (biomolecules). Some of these molecules can be produced in the absence of biology (abiotic). There is no natural place on Earth where only one microorganism exists, microbes thrive within the balance of a microbial community (1). Therefore this investigation looked at the change in the overall microbial community, rather than a single organism. For this research we chose the microbial ecosystem of a salt crust, due to the already selective desiccation resistance within the community. Halophile ecosystems are models for life in extreme environments including planetary surfaces. Our research was on the microbial preservation potential of salt to Mars conditions. We report on changes to the salt crust photosynthetic community lipid content measured when exposed to one week of simulated martian conditions (UV, pressure, temp) in a Mars chamber. Phospholipid fatty acid (PLFA) analysis was employed to determine changes in microbial community from exposure to the Martian environment.

The microbial ecology of the salt crust - black, green, and pink stratified layers of photosynthetic organisms vary with depth as a function of environmental conditions (Figure 1). These layers were exposed to Martian Condition in the Mars Chamber under three experimental set-up parameters:

Set-up 1. Thin layer = ~1 cm of salt crust was spread in a petri dish then placed in the Mars Chamber.

Set-up 2. Thick Layer- Lid off~ 3 cm of salt was spread in a petri dish and the lid of the petri dish was placed back on, then the dish was placed in the Mars Chamber.

Set-up 3. Thick Layer- lid on~3 cm of salt was spread in a petri dish and the lid remained off, the dish was placed in the Mars Chamber.

Sample- Setup The Mars Simulation conditions (UV, temperature and pressure) were for 1 week on the surface. Sample 1 is a piece of the salt crust, not exposed to simulated Martian conditions. Sample 2 (set-up 1) are thinly dispersed layers of salt to determine if thickness was a major factor in microbial survival. Sample 3(set-up 2) is a 3cm thick layer of salt with the lid removed. Sample 4(set-up 3) is the salt in a 3 cm thick layer within the petri dish. The petri dish lids (quartz glass) remained on to simulate the shielding effects of dust.

Microbial Analysis using Phospholipid Fatty Acid (PLFA) was used to determine changes within the microbial community (2). PLFA method is a standard method used to study changes in the microbial community when changes in environmental physical and chemical conditions occur. The Lipids analyzed during PLFA are as follows: A. Monoenoic-abundant in proteobacteria and adapt quickly to a variety of environments, B. Mid-Chain Branched Saturated (MidBrSats), common in sulfate reducing bacteria. C. Normal Saturated (Nsats)- found in all organisms- indicates less diverse populations. And D. Polyenoic- found in eukaryotes such as fungi, protozoa,
and algae. In addition to lipid analysis cells per gram is determined based on a cell equivalents of the total amount of PLFA’s extracted.

Results: Sample 1: Natural salt crust- not exposed to high UV.
Cells/gram: 114, Monolipids: 27.69%, Mid-BrSats: 0.43%, NSats: 43.51%, Poly: 27.49%

Sample 2: Thin layer salt- the most exposed to high UV
Cells/gram: 155, Monolipids: 34.91%, Mid-BrSats: 0.47%, NSats: 39.85%, Poly:23.66%

Sample 3: Thick Layer exposed salt- with the lid off- the second most UV exposed sample.
Cells/gram: 123, Monolipids: 34.94%, Mid-BrSats: 0.53%, NSats: 41.95%, Poly:21.69%

Sample 4: Thick layer – with the lid on- the most protected of the UV exposed samples.
Cells/gram: 114, Monolipids: 32.62%, MidBrSats: 0.39%, NSats: 41.43%, Poly: 24.37%

Discussion:
The Lipid analysis reveals interesting changes in the microbial community. The sample that was the most exposed to the Mars UV, had an increase in cells per gram, an increase in mono lipids, a slight increase in MidBrSats a decrease in polys and a significant decrease in NSats compared to the natural sample. This suggests that the stress caused by UV was enough to trigger an EPS response in production of lipids for protection, specifically the mono-lipids- the simplest lipids. The reduction in Nsats suggests that one organism is dominating this stress response. The cells per gram is an artifact of the way the measurement is based on lipid content, the second sample increased the lipid production leading to an apparent increase in cells. Samples three and four were also stressed to begin EPS production, but not to the extent of sample 2. The community diversity was reduced, but not significantly reduced compared to the natural state. Results from this investigation lead to a better understanding of the microbial response to UV radiation and the dominant lipids employed by the organisms for protection and survival.


Figure 1: Salt crust showing the three layers of stratified photosynthetic organisms.