SURVIVAL OF HALOPHILIC ARCHAEA IN THE STRATOSPHERE AS A MARS ANALOG: A TRANSCRIPTOMIC APPROACH. S. DasSarma[1], P. DasSarma[1], V. Laye[1], J. Harvey[2], C. Reid[2], J. Shultz[2], A. Yarborough[2], A. Lamb[2], A. Koske-Phillips[2], A. Herbst[2], F. Molina[2], O. Grah[2] and T. Phillips[2], [1]Department of Microbiology and Immunology, Institute of Marine and Environmental Technology, University of Maryland School of Medicine, Baltimore, MD 21202 and [2]Earth to Sky Calculus and Spaceweather.com.

Introduction: Idenifying potentially habitable regions of Mars is of great signifcance. In this context, seasonal dark streaks or recurring slope lineae (RSL) recorded on the walls of Garni crater captured by NASA's Mars Reconnaissance Orbiter are of interest [1]. The occurrence of RSLs at subzero temperatures suggest salty brine flows seasonally on the Martian surface melted by freezing-point depression, which is also supported by spectroscopic evidence for hydrated sodium and magnesium chloride, chlorate, and perchlorate salts in the Phoenix lander site [2-4].

On Earth, brines are nearly ubiquitous and generally thalassic. They harbor a great variety of halophilic microorganisms originating from all three branches of life, Archaea, Bacteria, and Eukarya [5]. Halophilic Archaea are able to tolerate the highest salinity due to negatively charged proteins which remain soluble and compete successfully with ions for hydration [6]. Discovery of brine flows on the surface of Mars has intensified interest in the polyextremophilic character of halophilic Archaea in relation to astrobiology [7].

Halophilic Archaeal Models for Astrobiology: Among halophilic Archaea (Haloarchaea), *Halobacterium* sp. NRC-1 has been extensively studied for its polyextremophilic character [8]. This species is capable of tolerating multimolar concentrations of sodium and potassium chlorides, including perchlorates. NRC-1 is also slightly thermotolerant with optimum growth at 42 ºC and survival at 49-50 ºC [9] and is resistant to UV and ionizing radiation [10,11]. Genomic and transcriptomic studies have established a range of mechanisms operating in NRC-1, including highly acidic proteins, and direct photorepair, double-stranded gap repair, and nucleotide excision repair systems.

Halorubrum lacusprofundi is another Haloarchaeon relevant to astrobiology, which was isolated from Deep Lake in the Vestfold Hills of Antarctica [12]. Deep Lake is perennially cold, with the temperature remaining subzero for more than 6 months of the year. However, Deep Lake does not freeze, even when temperatures drop to -18 ºC due to freezing-point depression from high salinity. *H. lacusprofundi*, which is capable of growth down to -2 ºC, is well-adapted to this environment. *H. lacusprofundi* biofilms have been reported as a possible mechanism for enhanced survival at the lowest temperatures. The *H. lacusprofundi* genome has been sequenced, a DNA microarray developed, and its proteins analyzed for cold activity, reduced surface acidity, and enhanced internal flexibility [13,14].

Haloarchaeal Models in the Stratosphere: Earth's stratosphere exhibits multiple extremes, including cold temperatures, high radiation, and low pressures, similar to those found on the surface of Mars [15]. In order to determine whether *Halobacterium* sp. NRC-1 and *H. lacusprofundi* may survive such extreme conditions, we launched live cultures of the mesophilic model *Halobacterium* sp. NRC-1 and the cold-adapted Antarctic isolate *Halorubrum lacusprofundi* into Earth's stratosphere on helium balloons. After return to Earth, the cold-adapted species showed nearly complete survival while the mesophilic species exhibited only slightly reduced viability.

Parallel studies in the laboratory showed that the cold-adapted species was better able to survive due to superior tolerance to freezing and thawing. Finally, genome-wide transcriptomic analysis [6] was used to compare the two haloarchaea at optimum growth temperatures versus low temperatures supporting growth. The cold-adapted species displayed perturbation of a majority of genes by cold temperature exposure, divided evenly between up-regulated and down-regulated genes, while the mesophile exhibited perturbation of only a fifth of genes, with nearly two-thirds being down-regulated. These results point to the importance of a regulation of a large number of genes in the cold-response of *H. lacusprofundi* likely important for survival in the stratosphere.

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