

**CAN TERRESTRIAL MICROBES GROW ON MARS?** W. L. Nicholson, Dept. of Microbiology & Cell Science, University of Florida, Space Life Sciences Laboratory, 505 Odyssey Way, Merritt Island, FL USA 32953. E-mail: WLN@ufl.edu.

**Introduction:** A central goal of Astrobiology is to explore the limits at which life can occur and to search for life and habitable locations outside Earth. Mars is currently an active target in the search for life due to its relative proximity and similarity to Earth, coupled with increasing evidence pointing to the past and present existence of liquid water at the surface and near subsurface [1]. Exchange of rocky impact ejecta between Mars and Earth has been known for at least two decades [2], and evidence has accumulated supporting the hypothesis that living microorganisms embedded in rocks could survive the transfer process [3]. Understanding the ability of terrestrial microorganisms to grow in the near-surface martian environment is of prime importance both for life detection and for protection of Mars from forward contamination by human or robotic exploration [4].

The surface environment of Mars presents formidable challenges to life, such as: harsh solar radiation; a scarcity of liquid water and nutrients; extreme low temperatures; and a low-pressure, CO<sub>2</sub>-dominated anoxic atmosphere [5]. Recent work in our laboratory has concentrated on investigating the possibility that prokaryotes from Earth could either (i) live on Mars in their current form, or (ii) evolve the ability to live under Mars conditions. Our experiments have involved environmental chambers that can simulate Mars atmospheric conditions of low pressure (P; 0.7 kPa), temperature (T; 0°C), and a CO<sub>2</sub>-dominated anoxic atmosphere (A), called here collectively low-PTA conditions.

**Growth of permafrost bacteria under low-PTA conditions:** Because much of the water on present-day Mars exists in a permanently frozen state mixed with mineral matrix, terrestrial permafrosts are considered to be analogs of the martian environment [6]. We therefore screened Siberian permafrost soils for microbes capable of growing under low-PTA conditions. Using this approach we reported the isolation of 6 *Carnobacterium* spp. isolates from Siberian permafrost that were capable of low-PTA growth [7]. In addition, a laboratory strain of *Serratia liquefaciens* was also found to grow under low-PTA conditions [8].

**Evolution of *Bacillus subtilis* to growth at low-P:** Previous work had indicated that most bacteria, including the common laboratory strain of *B. subtilis*, were unable to grow at pressures lower than ~2.5 kPa [9]. To investigate if *B. subtilis* could evolve the ability to grow at low-P, we cultivated strain WN624 at the

near-inhibitory pressure of 5 kPa for 1,000 generations, and isolated a low-P adapted strain designated WN1106 [10]. In competition experiments the low-P adapted strain showed higher relative fitness than the ancestor at 5 kPa, but not at normal Earth pressure (~101 kPa) [10]. Transcription microarray analyses indicated that exposure to low-P induced both the *sigB*-mediated General Stress Response and the anaerobic response in both the ancestral and low-P adapted strains [11]. Whole genome sequencing revealed that low-P adapted strain WN1106 had accumulated 12 mutations not present in the ancestral strain. The significance of these mutations to low-P adaptation in *B. subtilis* is currently being explored.

**References:** [1] Chyba C.F. & Hand K.P. (2005) *Annu. Rev. Astron. Astrophys.* 43, 31. [2] Jagoutz E. (1991) *Space Sci. Rev.* 56, 13. [3] Nicholson, W.L. (2009) *Trends Microbiol.* 17, 243. [4] Nicholson W.L. et al. (2009) *Trends Microbiol.* 17, 389. [5] Schuerger A.C. (2004) In *Martian Expedition Planning*, ed Cockell C.S. (Univelt Publishers), 363. [6] Frolov A.D. (2003) *JGR-Planets*, 108(E4), 7. [7] Nicholson W.L. et al. (2013) *Proc. Natl. Acad. Sci USA* 110, 666. [8] Schuerger A.C. et al. (2013) *Astrobiology* 13, 115. [9] Schuerger A.C. & Nicholson W.L. (2006) *Icarus* 185, 143. [10] Nicholson W.L. et al. (2010) *Appl. Environ. Microbiol.* 76, 7559. [11] Waters S.M. & Nicholson, W.L. (2013), submitted.

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