INSIGHTS FROM CYANOBACTERIAL GENOMES FOR THE DEVELOPMENT OF EXTRATERRESTRIAL PHOTOAUTOTROPHIC BIOTECHNOLOGIES. I.I. Brown[1], D.A. Bryant[2], S.G. Tringe[3], K. Malley[4], O. Sosa[4], S.A. Sarkisova[1], D.H. Garrison[1], D.S. Mckay[5]. [1]SARD/JSC, Mail Code: JE 23, ESCG, P. O. Box 58447, Houston, TX,. [2]The Pennsylvania State University, University Park, PA 16802, [3]DOE Joint Genome Institute [4] NASA/USRP, [5]NASA JSC.

Introduction: In-situ production of consumables (mainly oxygen) using local resources (In-Situ Resource Utilization-ISRU) will significantly facilitate current plans for human exploration and settlement of the solar system, starting with the Moon.

With few exceptions, nearly all technologies developed to date have employed an approach based on inorganic chemistry [1]. None of these technologies include concepts for integrating the ISRU system with a bioregenerative life support system and a food production system.

Therefore, a new concept based on the cultivation of cyanobacteria (CB) in semi-closed biogeoreactor, linking ISRU, a biological life support system, and food production, has been proposed [2, 3]. The key feature of the biogeoreactor is to use lithotrophic CB to extract many needed elements such as Fe directly from the dissolved regolith and direct them to any technological loop at an extraterrestrial outpost [4]. Our studies showed that siderophilic (Fe-loving) CB are capable to corrode lunar regolith stimulants because they secrete chelating agents and can tolerate [Fe] up to 1 mM [5].

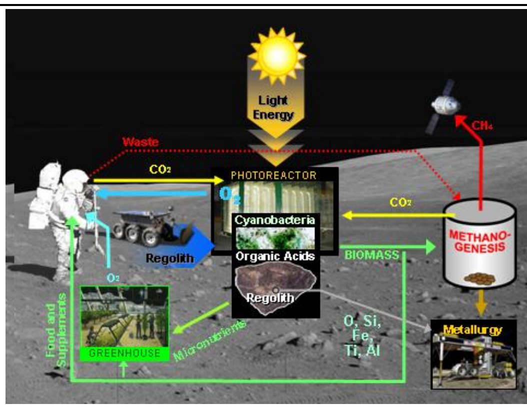

Fig. 1. Author's concept of a biotechnological loop for in situ resources extraction, propellant and food production at the lunar outpost

However, lunar and Martian environments are very hostile (very high UV and γ-radiation, extreme temperatures, deficit of water). Thus, the selection of CB species with high potential for extraterrestrial biotechnologies that may be utilized in 15 years must be sponsored by NASA as soon as possible. The study of the genomes of candidate CB species and the metagenomes of the terrestrial environments which they inhabit is critical to make this decision.

Here we provide preliminary results about peculiarities of the genomes of siderophilic CB revealed by analyzing the genome of siderophilic cyanobacterium JSC-1 and the metagenome of iron depositing hot spring (IDHS) Chocolate Pots (Yellowstone National Park, Wyoming, USA).

Methods: The amino acid sequences of proteins involved in the maintenance of Fe homeostasis and oxidative stress in several species of both siderophilic and non-siderophilic CB were used as anchors to find the proteins in the JSC-1genome and metagenome of Chocolate Pots.

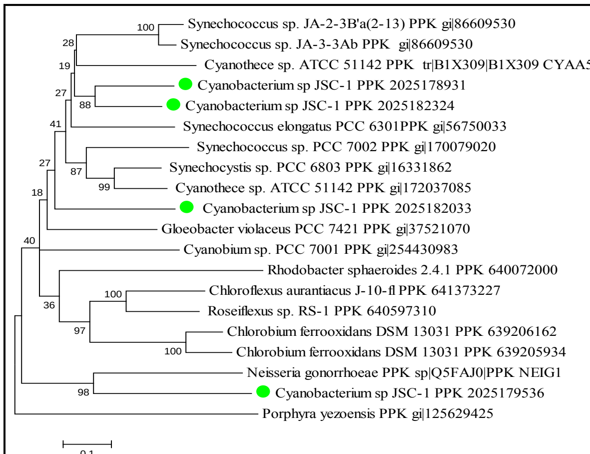

Fig. 2. Phylogenetic analysis of the Polyphosphate kinase (PPK) proteins in siderophilic CB JSC-1using the Neighbor-Joining method [6]. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (500 replicates) are shown next to the branches [7]. Distances were computed using the Poisson correction method [8] and are in the units of the number of amino acid substitutions per site. Phylogenetic analyses were conducted in MEGA4 [9].

Results: Siderophilic CB JSC-1 has been identified only in LaDuke IDHS to date but not in any other hot spring with low [Fe] (< 1 μ). Analysis of the JSC-1 genome revealed that this siderophilic species possesses 2 proteins belonging to the Bacterioferritin (Bfr) domain, 4 proteins belonging to DNA-binding ferritin-like protein (Dps) super family and 4 proteins belonging to polyphosphate kinase (PPK) family. CB *Synechococcus* sp. JA-2-3B'a (2-13), which inhabits hot springs with low and high [Fe], possesses 1 protein belonging to Bfr domain and 1 PPK but 3 proteins belonging to Dps superfamily. In contrast, such non-siderophilic CB as *Synechocystis* sp. PCC 6803 and *Synechococcus* sp. PCC 7002 possess' only 1 representative of proteins belonging to Bfr domain, Dps superfamily and PPK family.

Conclusion: It has been shown that IDHS are richer with ferrous iron than the majority of hot springs around the world [10, 11]. Fe^{2+} is known to increase the magnitude of oxidative stress in prokaryotes through so called Fenton reaction [12, 13]. It is not surprising, therefore, that the CB inhabiting IDHS have larger sets of the proteins involved in the maintenance of Fe homeostasis and oxidative stress protection than non-siderophilic CB. This finding combined with our earlier results about the ability of some siderophilic CB to utilize chemical elements released from analogs of lunar and Martian regolith [4] make them the most advanced candidates to be employed in advanced extraterrestrial biotechnologies.

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