

Progress towards a meta-database of spacecraft-associated microorganisms.

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Introduction:

The microorganisms associated with spacecraft and related surfaces represent the primary pool of forward contaminants that may compromise the integrity of future life-detection missions [1, 2]. Herein, we describe the construction of a meta-database of these microorganisms that may ultimately serve as a bioinformatics-type resource for the space science community. Using student-centered efforts, the Spacecraft-Associated Microbial Meta-database (SAMM) is being assembled using information retrieved from the public domain, including scientific reports and the ESA/DSMZ culture collection. Presently, SAMM is publically available at the following site (<http://www.cpp.edu/~rmogul/cpp/SAMM.html>) and includes over 1400 entries from NASA and ESA-related facilities and all domains of life, where the microbial entries are organized around 33 meta-tags that relate to taxonomy, Gram stain, spore forming properties, habitat, source of isolation, location of isolation, cleanroom class, types of characterization, and growth conditions.

Undergraduate Education Component:

The construction of SAMM is an undergraduate undertaking conducted as part of an ongoing research-based course in the College of Science at Cal Poly Pomona. The academic objectives of this capstone-like course are to improve fluency in the biological sciences and strengthen critical analysis skills by challenging students with the deconstruction and interpretation of interdisciplinary scientific articles that combine space sciences with modern techniques and principles in microbiology, molecular biology, genetics, and ecology.

Preliminary Analyses:

Holistically, SAMM currently contains 1446 microorganisms and includes 952 bacterial, 62 archaeal, and 118 fungal entries obtained from 11 different scientific articles and 157 bacterial entries obtained from the DSMZ collection. In terms of taxonomy, only 26% of the entries were cultivated and subsequently characterized, whereas 84% were characterized *sans* cultivation using molecular genetics techniques.

For the bacterial entries possessing Gram stain metadata (945), the analyses show that 61% are Gram-positive, 38% are Gram-negative, and ~1% are variable. Analyses at each spacecraft-associated facility further demonstrates the abundance of Gram-positive bacteria, as ~60-80% of the entries from the Kennedy Space Center (KSC), Johnson Space Center (JSC), Lockheed Martin (LM), European Space Research and Technology Centre (ESTEC), and European Aeronautic Defense and Space Company (EADS) are Gram-positive. In contrast, the bacterial entries from the Jet Propulsion Laboratory (JPL) are mostly Gram-negative at 52%, with 48% being Gram-positive.

When considering the spore forming metadata for bacteria from all facilities (659 entries), 58% and 42% of the

entries are listed as non-spore forming and spore forming, respectively. The dominant abundance of non-spore forming bacteria is illustrated by the bacterial entries from JPL and KSC, where 67 and 75% are non-spore forming (and 33 and 25% spore forming), respectively. When considered in the context of Gram-staining, our analyses indicate that the composition of SAMM is ~40% spore-forming and Gram-positive bacteria, ~27% non-spore forming and Gram-negative bacteria, and ~22% non-spore forming and Gram-positive bacteria.

The diversity among the spacecraft-associated bacteria was measured at both the phylum and genus levels. These studies show that the Firmicutes (40%), Proteobacteria (33%), and Actinobacter (16%) are the most abundant phyla across all facilities (1083 entries). However, when comparing across JPL (493) and KSC (365), very notable differences in phylogenetic breadth are observed. For instance, the bacterial phyla at JPL are significantly more diverse, as 21 phyla are associated with JPL and only 9 phyla are associated with KSC.

Analyses of the bacterial genera metadata (680 entries) show that the *Bacillus* (39%), *Acinetobacter* (6%), and *Staphylococcus* (5%) are the most abundant across all facilities. When comparing across JPL (133) and KSC (321), opposite trends in diversity of genera and phylum are observed. For example, the bacterial genera at KSC are significantly more diverse, as 80 genera are associated with KSC and only 21 with JPL. Among these genera, the *Bacillus* (26%), *Ralstonia* (5%), and *Acinetobacter* (4%) are the most abundant at KSC; while for JPL the *Bacillus* (26%), *Staphylococcus* (20%), *Propionibacterium* (9%) and *Acinetobacter* (4%) are the most abundant. These nested comparisons (using the current state of SAMM) also show a unique geographical distribution for many genera where, for example, *Aquabacterium* and *Oceanobacillus* are only associated with KSC, whereas *Alcaligenes* and *Propionibacterium* are only associated with JPL.

Conclusions and Next Steps:

The ultimate aim of this endeavor is to construct a publically accessible compendium of spacecraft-associated microorganisms that will serve as a bioinformatics resource for the space science community. To that end, our long-term plans for SAMM involve the inclusion of microbial inventories from multiple space-faring agencies, as well as those from the JPL catalogs, which contain microorganisms not listed in the public domain. In this presentation, therefore, the trends learned from the analysis of SAMM and future crowd sourcing efforts will be discussed.

References:

- [1] Space Studies Board (2006) *Preventing the Forward Contamination of Mars* National Academies Press, Washington DC.
- [2] La Duc et al. (2014) *Astrobiology* 14:15-23.