THE VIABLE MICROBIOME OF SPACECRAFT ASSEMBLY CLEANROOM ENVIRONMENTS: METAGENOME OF THE LIVING. Parag Vaishampayan[1§], Thomas Weinmaier[2], Alexander J. Probst[3], Myron La Duc[1], Natalia Ivanova[4], Thomas Rattei[2]. ([1]Biotechnology and Planetary Protection Group, Jet Propulsion Laboratory, California Institute of Technology, Pasadena, CA 91109, USA. [2]Division of Computational Systems Biology, Department of Microbiology and Ecosystem Science, University of Vienna, Althanstrasse 14, 1090 Vienna, Austria. [3]Department of Earth and Planetary Science, University of California, Berkeley, 307 McCone Hall, Berkeley, CA 94720, USA. [4]DOE Joint Genome Institute, Walnut Creek, CA, USA. [§]Presenting author)

The inadvertent introduction of terrestrial microorganisms or other contaminants could seriously jeopardies the scientific integrity of the space mission and could compromise our ability to discriminate authenticity of Martian bio signature from a terrestrial organism. The National Research Council has recommended that "Uncertainties in the current assessment of Martian habitability and the potential for the inclusion of living entities in samples returned from Mars may be reduced by continued studies of the metabolic diversity and environmental limits of microbial life". It further suggests to develop molecular (DNA)-based identification techniques for mitigating the planetary protection risks (forward and reverse). For this reason, it is important to develop a comprehensive inventory of microbes that are present on spacecraft to avoid interpreting their traces as authentic extraterrestrial biosignatures. Culture-based methods are currently used by NASA to assess spacecraft bioburden but could detect only a very small subset of total organisms' present.

The findings of recent studies employing 16S rRNA gene sequencing and metagenomic analyses from sample-derived total DNA extracts have postulated a reciprocal dependency between indoor and human microbiomes. However, the results of this investigation demonstrate that the metagenome exclusive to viable cells differs significantly from that derived from the total DNA recovered from indoor environments. A molecular viability marker was applied to samples collected from a cleanroom facility, and subsequent metagenomic sequencing experiments showed considerable differences between the resulting viable-only and total microbiomes. The composition of the viable bacterial communities associated with uncontrolled gowning areas differed significantly from that of controlled cleanroom environments, implicating selective pressure on indoor bacteriomes by more stringent facility maintenance and cleaning. Nevertheless, analyses of sequence abundance suggested that the viable microbiome was influenced by both the human microbiome and the ambient ecosystem external to the facility, which resulted in a complex community profile. Differences in sequence abundance and functional capabilities between samples suggested a decrease of oxygen-dependent organisms in the cleanroom environment. Also detected were the first viral signatures ever retrieved from a cleanroom facility: the genomes of human cyclovirus 7078A and Propionibacterium phage P14.4. The findings presented here, as well as the innovative methods that enabled their discovery, promise to have profound implications on the design and interpretation of ongoing and future indoor microbiome studies.