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## MICROGRAVITY COMPATIBLE, PORTABLE, AND AUTOMATED NUCLEIC ACID EXTRACTION SYSTEM FOR FUTURE HUMAN MISSIONS

## Abstract

To perform omics in space, spacefaring nations need to develop instrumentations for extracting nucleic acids and sequencing methodologies for inflight detection and measurement of biomolecules related to physiological and immunological effects related to spaceflight (e.g., crew health, elevated antimicrobial resistance, and virulence). In order to determine if spaceflight induces changes in crew and environment aboard the human habitat, microgravity-compatible molecular microbiology instrumentations are essential. We have developed and validated the simulated micro()gravity tested instrument for automated nucleic acid extraction (Titan) system, to perform hands-free nucleic acid extractions in remote extreme environments, Yellowstone National Park. The Titan system is compact and extracted nucleic acids from most human samples, air, surface, and water.

Dr. Kathleen Rubins, a NASA astronaut, became the first person to sequence DNA on the ISS, using the Oxford Nanopore MinION sequencer. While this was a great milestone for space molecular biology, the DNA that was sequenced had already been extracted and the metagenome libraries prepared on Earth before sending to space for sequencing. In another project, bacterial DNA, which also had been isolated on Earth, was sent to the ISS and successfully amplified with the miniPCR. More recently, methods have been developed to allow for nucleic acid extraction to occur on the ISS before in-situ downstream analyses. For example, with the Genes in Space-3 project, cultured bacterial cells that had been isolated from around the ISS were lysed in a thermocyler and the DNA amplified before being sequenced on the MinION. With the WetLab-2 research platform, RNA was successfully isolated from Escherichia coli cultures by lysing the cells with bead beading and then capturing the released RNA with the RNA express<sup>TM</sup> column, after which the isolated RNA was analyzed with RT-qPCR. While the current methods that allow for sample-to-analysis entirely aboard the space station are a tremendous advancement for space genomics, these techniques are manual and take up crew time, are not high throughput and are better suited for pure cultures rather than complex, mixed microbial communities, as they are not efficient for low biomass samples and the nucleic acids are not pure due to the left-over cell debris. To address these limitations, we developed Titan, an automated sample processing system, that is microgravity compatible, needing minimal crew time, reducing the amount of contamination between samples, producing consistent results, and can process multiple complex samples simultaneously in an easy to use streamlined manner.