Successful Validation of Sample Processing and Quantitative Real-Time PCR Capabilities on the International Space Station

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The WetLab-2 system was developed by NASA Ames Research Center to offer new capabilities to researchers. The system can lyse cells and extract RNA on-orbit from different sample types ranging from microbial cultures to animal tissues. The purified RNA can then either be stabilized for return to Earth or can be used to conduct on-orbit quantitative Reverse Transcriptase PCR (qRT-PCR) analysis without the need for sample return. The qRT-PCR results can be downlinked to the ground a few hours after the completion of the run.

The validation flight of the WetLab-2 system launched on SpaceX-8 on April 8, 2016. On orbit operations started on April 15th with system setup and was followed by three quantitative PCR runs using an *E. coli* genomic DNA template pre-loaded at three different concentrations. These runs were designed to discern if quantitative PCR functions correctly in microgravity and if the data is comparable to that from the ground control runs. The flight data showed no significant differences compared to the ground data though there was more variability in the values, this was likely due to the numerous small bubbles observed.

The capability of the system to process samples and purify RNA was then validated using frozen samples prepared on the ground. The flight data for both *E. coli* and mouse liver clearly shows that RNA was successfully purified by our system. The *E. coli* qRT-PCR run showed successful singleplex, duplex and triplex capability. Data showed high variability in the resulting Ct's likely due to bubble formation and insufficient mixing during the procedure run. The mouse liver qRT-PCR run had successful singleplex and duplex reactions and the variability was slightly better as the mixing operation was improved.

The ability to purify and stabilize RNA and to conduct qRT-PCR on-orbit is an important step towards utilizing the ISS as a National Laboratory facility. The ability to get on-orbit data will provide investigators with the opportunity to adjust experimental parameters in real time without the need for sample return and re-flight.

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