PROJECT SUMMARY

Purpose of the research

The purpose of this Phase 1 development was to determine feasibility of a Rapid Coliform Detector (RCD) to detect the presence or absence of *E. coli* bacteria in a water sample, with a desired total test duration of 30 minutes. The ultimate objective of this research is to develop an RCD to detect and quantify contamination by *E. coli* bacteria in potable water on board the ISS and other space exploration habitats and provide results in approximately 30 minutes.

Brief description of the research carried out

The Phase 1 research included development of functional requirements for a ground-based prototype RCD and baseline requirements for a spaceflight RCD. The chemistry for amplification and detection of *E.coli* rRNA was developed, along with the chemistry and method of freeze-drying the amplification reagent in a solid form for long-term storage. Solid phase extraction chemistry was investigated to determine potential to remove inhibitors. Several iterations of a prototype design were developed, and a prototype amplification/detection cartridge was fabricated and assembled. Many aspects of the proposed amplification/detection process were tested, some with the prototype and some with lab equipment. Designs and test results were evaluated and a conceptual design of the spaceflight RCD cartridge and support hardware was developed.

Research findings or results

All Phase 1 objectives were achieved:

- Requirements for ground test hardware and spaceflight hardware were developed.
- PyroScript detection system for *E. coli* rRNA was designed.
- Isothermal amplification chemistry was developed.
- Prototype test kit cartridge was designed and fabricated.
- Test methods and the prototype test kit cartridge were evaluated.
- Conceptual design of spaceflight test kit cartridge and process controller was completed.

Tests showed that the chemistry that was developed will successfully amplify and detect *E. coli* rRNA in less than 30 minutes in a lab setting, with no false-positive readings, and a shelf-life of at least ten weeks. It was determined that passing the sample water through an adsorbent at the start of the process may increase speed of the reaction, but is not essential to successful detection. Designs were developed to consolidate all the functional requirements of the laboratory equipment into a single cartridge. *E. coli* was successfully detected in a water sample using the prototype hardware that was developed. A conceptual design of a spaceflight RCD system showed that the disposable cartridge can have a total mass as low as 9.0 g and the supporting control hardware can have a mass of approximately 1.3 kg.

Do the results justify Phase II continuation?

Results of the chemistry development tasks show that amplification and detection of *E. coli* DNA or RNA in a laboratory setting can be performed accurately and reliably. System design and development tasks have shown significant progress toward translating the proven laboratory conditions and operations to a spaceflight hardware design. The effect of gravity on fluid flow in the RCD does not appear to be a factor in our system design and the primary challenges appear to be thermal performance and control, and these challenges can be achieved. Prototype testing of an RCD cartridge in Phase 1 led to many lessons learned and design improvements for the next phase of development: these improvements are incorporated into the conceptual spaceflight design. Additional development and testing of a complete RCD system prototype, including the controller, will enable refinement of designs and operations that will result in a stand-alone RCD system that yields results in space that are similar to the current laboratory procedures.