Rapid and Simple Sample Acquisition During Space Flight:

Simultaneous Extraction of Proteins and Nucleic Acids from Bodily Fluids and Cabin Water Using Free-Flow **Bi-directional Isotachophoresis**

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Research Objectives

- The overall goal of this proposal is to develop molecular pre-concentration, separation, extraction and amplification devices for proteins and nucleic acids for integration with in situ instrument systems in flight missions.
- ٠ We will develop an electrophoresis method for extracting both proteins and nucleic acids from a single fluid sample using a new type of free-flow bi-directional isotachophoresis (ITP).
- ٠ The innovation is an easy-to-use, portable, and lightweight fluidic platform capable of rapidly extracting, concentrating, and separating proteins and nucleic acids simultaneously from a liquid sample for integrated use with downstream in situ flight instruments.
- Current sample extraction methods are not ideal for space crew with limited time • and resources, as they require long processing times, are labor intensive, and often use hazardous chemicals. The project is TRL 1. We have demonstrated

that free-flow ITP in simple fluidic devices is

possible. We have not initiated research to

extract and separate proteins or nucleic

acids. We plan on translating our work to

extract target molecules from real world fluid samples. Funds for the proposed effort

The success of the project will dramatically

capabilities with a molecular extraction and

amplification method that is revolutionary

improve current sample acquisition

in ease-of-use and flexibility.

will be used to elevate this project to TRL 3.

Anionic Leading Electrolyte Fluid Sample + Terminal Electrolyt (Bodily Fluid, Cabin Water) **Bi-directional** Protein and Nucleic Acid Isotachophoresis Targeted Protein Cationi eading Electrolyte **Potential Impact** Electric Field Unwanted Molecules Nanocomposite Electrode

Approach

My lab has developed small simple fluidic devices with nanocomposite electrodes that perform free-flow ITP separation and concentration of charged molecules and proteins. Further technical development is required to exploit our devices for sample acquisition during space flight missions.

Building on our previous microfluidic expertise, we will design and fabricate new freeflow ITP devices and investigate the electrophoresis behavior of nucleic acids and proteins in "clean" controlled mixtures of synthetic proteins and nucleic acids. The next step in our approach will be to investigate the ability to extract both nucleic acids and proteins simultaneously from a single sample using bi-directional ITP. We will combine mixtures of nucleic acids and proteins. Mixture of sample will driven into our ITP devices and we will investigate the separation and concentration efficiency for a given applied voltage, flow rate, sample pH, and buffer chemistry. With an improved understanding of how nucleic acid and protein mixtures impact ITP performance, we will then focus our efforts on developing the ability to extract target proteins and nucleic acids from real world fluids.

Because separation, concentration and extraction of both nucleic acids and proteins will be accomplished in a single step, the proposed bidirectional ITP sample purification system offers an unprecedented level of operational simplicity. The system will require only small volumes of reagents and enable significant improvement in existing space science abilities as extracted samples will be capable of being used with downstream in situ instruments. The proposed research will also dramatically improve sample purification and enrichment capabilities for existing meso- and microfluidic systems. The ITP hardware will be compatible with not only analytical instruments, but also with many existing microfluidic systems and devices. This project therefore has the potential to be crosscutting and serve as a valuable genetic and proteomic preparatory tool for fluidic applications in detection and prevention of disease in developing countries, biowarefare/anti-terrorism applications, environmental monitoring, point-of-care diagnostic testing and for basic biological research.