CHIPPS: CHARGED INFORMATION-STORAGE POLYMER PREPARATION SYSTEM. K.F. Bywaters[1], A.J. Ricco[2], T.D. Boone, T. Hoac, R.C. Quinn[2], K. Zacny[1] [1]Honeybee Robotics, 2408 Lincoln Ave, Altadena, CA 91001 [KFBywaters@honeybeerobotics.com; KAZacny@honeybeerobotics.com], [2]NASA Ames Research Center, Moffett Field, CA 94035 [antonio.j.ricco@nasa.gov; trinh.hoac@nasa.gov; travis.d.boone@nasa.gov; richard.c.quinn@nasa.gov]

Introduction: The search for life beyond Earth should incorporate technologies capable of detecting an array of potential biomarkers: this increases the chances of unambiguous results for autonomous missions to Ocean Worlds, such as Europa and Enceladus as well as Mars. Charged polymers comprised of multiple block types are biological systems' means to store and transfer information; life on other worlds may use different polymers than those of terrestrial biology. The examination and characterization of both mineralogical and biological charged particles ranging in size from 20 nm – 5 μm would detect a range of potential biomarker particles as one relevant part of an overall life-search instrumentation suite. The successful development of a synthetic nanopore Coulter Counter sample processing system will further detection capabilities for biomarkers beyond Earth.

Technological advances are required to achieve principal science objectives of a proposed Europa lander mission and multiple proposed Enceladus missions that will search for biosignatures of past or extant life. For life-detection missions, detection sensitivity and reliability are of particular concern due to small sample sizes (2 μL – 1 mL) and the extraordinary import of the results. The preparation and processing of small samples can constrain limits of detection (LoDs), as on the Viking [1] and Phoenix [2] landers and Mars Science Laboratory rover [3]. As we prepare to explore ocean-world environments, technological advances in autonomous sample processing will add new classes of reliably detectable biosignatures while simultaneously improving their LoDs.

Concept: The Charged Information-storage Polymer Preparation System (ChIPPS) will integrate a monolithic, microfluidic sample processor that prepares (simulated) icy-world samples for use with two complementary nanopore-based analyzers: 1) Oxford Nanopore's MinION - a charged-polymer sequencer that characterizes chain size, shape, and charge vs. chain position [4]; 2) Ontera's nano/microparticle sizer-and-counter, i.e. a Coulter counter, which characterizes the relative abundance of charged polymer chains (including fragments thereof) and other particles according to their hydrodynamic diameter, length, and charge [5-6]. Such measurements will reveal the nature and abundance of charged polymers that could be used by biological systems to store and transfer information—much as DNA and RNA are used terrestrially—without limitation to those terrestrial examples, given that life elsewhere may utilize different information store-and-transfer moieties. This key component of a search-for-life strategy must enhance the concentrations of target polymers sufficiently according to the platform: 400 ng DNA/equivalent for Oxford Nanopore's MinION (per manufacture's recommendations), 1 – 5 nM of DNA/equivalent for Ontera's polymer sizer/counter (per manufacture's recommendations). It must also adjust sample salt concentration to attain scientifically useful LoDs. These functions are particularly critical for an Enceladus fly-by sample capture, expected to be just a few μL of ice plume particles [7].

Technology Overview: Two critical functional processes to attain adequate LoDs for small volumes of likely-dilute ocean-world samples will be concentration of the extracted sample and the removal of excess salt. Our previous work focused on combining sample concentration via water vapor removal through a hydrophobic membrane [8, 9] with osmosis-driven dialysis to remove salt, both functions imple-

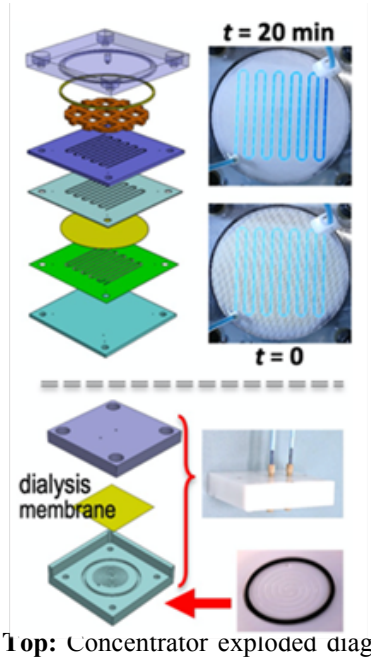

Figure 1. Top: Concentrator exploded diagram (left) and photos of concentration of blue dyed solution (right). **Bottom:** Dialyzer exploded diagram (left); photo of spiral microchannel (below right) and assembled dialyzer unit.

mented in fluidic-manifold-compatible format. Figure 1 shows that system's key components.

Multiple channel layouts, membrane areas, and internal support structures were developed and optimized for the concentrator; multiple types of dialysis membrane, pressures, flow rates, and channel depths were characterized for the dialyzer. Ultimately, both components functioned successfully, the former concentrating samples 10- fold in 60 min and the latter reducing salt concentration 10-fold in 75 min. Our results for model DNA samples are in preparation for publication [10]. These components will be optimized and integrated into the ChIPPS manifold.

Technical Approach: We build on our foundation of successful biology CubeSat payloads [11-18] and *BioSentinel* (delivery 2021) [19], along with WetLab-2 on the International Space Station [18] and experience with FLuidics for Ocean Worlds (FLOW) [11], to design, build, integrate components, and test the ChIPPS manifolded fluidic system. Fluidic components are selected from those flown in CubeSat and other missions [11-20], as well as those developed in the past three years by six NASA-funded projects that included manifold-based fluidic systems and interfaces for icy-world biomarker analysis [8 -9; 20]. Manifold design will apply lessons learned from those projects for component layout, tolerancing, mechanical fastening, and leak-tight sealing.

An abbreviated operational sequence begins with pre-evacuation of the manifold and priming with ultra-high-purity water; reconstituting the various dried reagents; preparing the binding column for charged polymers by rinsing/conditioning with salt solutions; drawing in sample, delivering it to the lyser, adding lysis buffer, then physically disrupting it; filtering (0.45 μm) sample lysate, mixing in binding buffer, then pumping to the ion-exchange column. The first elution of charged polymers is delivered to the dialyzer to reduce salt to ~ 0.05 M; it is then sent to the concentrator (~20x). About 100 μL of processed sample, matching analysis volume without waste, is sent via bubble trap to the nanopore analytical instruments. The second fraction is similarly eluted from the column and processed, followed by the third. With the configuration of Figure 2, all three eluted fractions can be concentrated ~20x without risk of NaCl precipitation. The ChIPPS project will evaluate a modified configuration to provide improved desalination, yielding 100 μL for nanopore analysis without exceeding the solubility limit of NaCl, whether for sequencing or sizing, to measure the lowest possible starting charged polymer concentrations.

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