A NEW APPROACH TO SENSITIVE ANALYSIS OF RESILIENT BIOMATERIALS. B. L. Henderson[1], V. Abrahamsson[1], J. Prothmann[3], F. Zhong[1], Y. Lin[1], W. Schubert[1], F. Chen[1], A. J. Williams[2], and M. Tuite[1]

[1]NASA Jet Propulsion Laboratory, Caltech Institute of Technology (4800 Oak Grove Dr., Pasadena, CA, 91109; Bryana.L.Henderson@jpl.nasa.gov), [2]University of Florida (Gainesville, FL), [3]Formerly JPL/Caltech, now German Federal Institute of Hydrology (Coblenz, Germany)

Introduction: The 2023-2032 Planetary Science and Astrobiology Decadal Report has reaffirmed the need for life-detection and organic-characterization strategies for understanding the evolution and habitability of target bodies across our solar system. To date, organic analysis instrumentation on previously-flown payloads has been hampered by low sensitivity and difficulties in distinguishing native organic signals from contamination or competing reactions induced by their high-temperature analyses.

Over the past decade, our lab has been addressing these challenges to provide a new option for planetary in-situ analysis. We have developed and miniaturized a new instrument with reduced complexity, lower-temperature extractions, and improved sensitivity, and have removed the risks inherent to other approaches by eliminating the need for organic solvents and derivatization. Our instrument, called the Supercritical CO_2 and Subcritical H_2O ANalysis instrument (SCHAN), uses only carbon dioxide and water to extract and analyze a wide range of organics and biomolecules (including chiral species) down to parts per trillion levels [1,2]. Here, we demonstrate that these solvents provide several notable advantages for biosignature analysis, and present some of our recent work on protein hydrolysis and cell lysis with these solvents.

Methodology: Subcritical water and carbon dioxide are known for their excellent extraction properties such as low viscosity and improved extraction rates, and are also able to lyse hardy cells and break down large biopolymers into more easily characterized subunits [3]. In this work, we use supercritical CO_2 to expose cell contents and carbonated subcritical water to increase the yield of the biomolecules being analyzed. Following this step, the SCHAN instrument extracts and preconcentrates the organics before sending them through a chromatography column and into a mass spectrometer for analysis.

Results and Discussion: We found that the pressurization of aqueous samples with CO_2 prior to heating acidifies the sample and significantly increases the rate of reaction and efficiency of protein hydrolysis. We have also recently determined that, when pressure and temperature is optimized, SCHAN can release biosignatures from biological cells (including *E. coli, B. atrophaeus,* and *Chlorella vulgaris*) at levels of 1×10^4 cells/mL or lower. This detection limit is lower than the 3×10^6 cells/mL reported for Viking's GC-MS on Mars [4].

Cell-lysing and protocols such as these that make biosignatures more easily accessible are needed to improve the odds of detection of life by chemical analysis techniques. Hardy materials may be difficult to identify without some type of pretreatment to release their contents – and a large intact biological protein (having hundreds or thousands of amino acid subunits) may be undetectable on its own. Releasing their contents for analysis increases the proportion of detectable biosignatures. Our work demonstrates that the simple solvents CO_2 and H_2O combined with heat and pressure can increase the number of detected biosignatures (e.g., fatty acids and amino acids) and cells by 1-2 orders of magnitude over untreated protein and cell samples.

Future Applications: SCHAN's sensitivity and versatility make this approach valuable for any mission where sensitive organic analysis or life detection is needed. Efforts to mature and miniaturize this system are ongoing, and environmental testing for Ocean World environments is currently underway.

Acknowledgments: This work was carried out at the Jet Propulsion Laboratory, California Institute of Technology, under a contract with the National Aeronautics and Space Administration (80NM0018D0004), and was supported through internal funding from JPL.

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