NASA Astrobiology Early Career Collaboration Award Report

## Biotic and Abiotic Organic Signatures Present in a 3.1 km Deep Brine with Implications for Subsurface Life on Mars

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Throughout July and August of 2019 I went on a six week field expedition to the Witswatersrand Basin in South Africa to explore the limits of microbial life in the deep continental subsurface. For the majority of my stay, I resided in a small mining town called Orkney from which I was able to drive to my field site, the gold and uranium mine Moab Khotsong. With the guidance and help of our mine contact Bennie Liebenberg, I was able to collect water, gas, and filter samples from three different fracture fluid reservoirs in the mine, at depths of 1.2km, 2.9km, and 3.1km. From the shallow site (1.2km) I gathered microbiology and geochemistry samples from a moderate and brackish fluid while at the deeper sites (2.9km, 3.1km) I was able to obtain such samples from high temperature, hypersaline brines.



When I was not sampling fluid filled fractures at the mine, I spent my time in Bloemfontein, South Africa both preparing equipment for sampling, and ensuring collected samples were stored correctly. This project provided me invaluable field experience in collecting subsurface fracture fluid samples, and with managing a large and diverse inventory of samples.

Over the six week period, my advisor Dr. TC Onstott (Princeton University) and each of the following collaborators assisted me at some point with the sampling and sample prep: Dr. Thomas Kieft (New Mexico Institute of Technology), Dr. Oliver Warr (University of Toronto), Dr. Errol Cason (University of the Free State), Dr. Julio Castillo Hernandez (University of the Free State), Dr. Jan-G

Vermeulen (University of the Free State). Crucial on ground project and sample planning support was provided by Dr. Esta van Heerden (North-West University) and equipment assistance was provided by Dr. Raymond J Durrheim (University of the Witwatersrand).

Since returning back to Princeton University, I have quantified the dissolved organic carbon (DOC) and dissolved inorganic carbon (DIC) fractions of my brine samples, as well as their  $\delta^{13}$ C values to find a large, light DOC pool and small, heavy DIC pool. Additionally, I have run IC-MS analyses on the brine organic fractions to find a decent proportion is due to low molecular weight compounds. Qualitative GC-MS results from my collaborator Dr. Cliff Walters (Exxon Mobil) and volatile hydrocarbon characterization from my collaborators Dr. Barbara Sherwood Lollar (University of Toronto) and Dr. Oliver Warr (University of Toronto) show the presence of many sulfur containing compounds and a potentially abiogenic formation signature in my volatile organic carbon pool. I have started exploring the biotic environment by assembling bacterial and archaeal genomes from metagenomic sequence data I have obtained on the shallow, brackish fluid, and am seeing evidence for sulfur and carbon cycling. So far, it appears I have a unique suite of carbon compounds and aqueous geochemistry in my brines that may support a microbial carbon cycling network that hasn't been seen before in brackish fracture fluids of the Witswatersrand Basin, and that might be similar in geochemistry to subsurface liquid environments that could be found on Mars. Recently, I was able to present these preliminary findings at the AGU conference in San Francisco, California.

In the next few months, I will continue my research by further characterizing the compounds in my brine DOC pool, including analysis of amino acids, and by obtaining gas samples for the volatile sulfur compounds so their isotopic signatures can be determined.  $\delta^{13}$ C values for the organic acids and phospholipid fatty acids will be determined with help from our collaborator Dr. Kate Freeman and lab at Penn State University. Additionally, I will be attempting extractions from the brine samples in order to characterize any inhabiting microbial communities.

Not only has this NASA early career collaboration award supported project been a great adventure, but it has allowed me to expand both my scientific skill set and understanding of microbial life and geochemistry in extreme environments. I am excited to build upon the unique and interesting results I was able to obtain through this field expedition as I continue my thesis research.