

Isolation of Cellulolytic Bacteria from High pH Serpentinizing Springs in the Philippines. B. Vallalar¹ and D. Meyer-Dombard¹, ¹University of Illinois at Chicago (uvalla2@uic.edu), (drmd@uic.edu).

Introduction: Serpentinizing ecosystems have previously been suggested as analogs for potential extraterrestrial habitats and the origin of life [1]. The process of serpentinization involves the hydrologic alteration of ultramafic rocks formed in Earth's mantle. The reaction between water and iron-rich minerals contained in these rocks produces hydrogen and methane gas, conducive for microbial metabolisms in these environments. The resulting fluid from these interactions is highly reduced and alkaline, and may breach the surface in various seeps found around the world. One primary focus of studying these environments involves the sources and cycling of carbon. Dissolved inorganic carbon is extremely limited in serpentinizing fluids, while organic carbon is more readily available in the form of methane and small quantities of other hydrocarbons [1]. Assuming life originated in serpentinizing environments, interpretations could then be made as to the adaptation of these initial communities to modern serpentinizing systems. A major difference in modern versus ancient systems would be the emergence of land plants around the beginning of the Ordovician period [2]. This external carbon source likely caused a shift in the surficial community composition in favor of different heterotrophic metabolisms. Specifically, utilization of cellulose is of interest as many of the springs in our field sites are found within heavily vegetated locations.

Field Site: Samples of serpentinizing and hydrothermal fluids and sediments were collected from ophiolites in the Zambales and Palawan regions of the Philippines. Ophiolites are geologic features in which deep mantle rocks have been uplifted to the surface, and may then be subject to hydrologic alteration. They therefore serve as terrestrial analogs to deep marine serpentinizing systems. The pH values for the fluids at the sites range from 10 - 12, and the average temperature of the fluid near the source was ~33°C. The springs are in densely vegetated areas with large quantities of dead and decaying plant matter added to the source pools.

Objective/Methods: The focus of this study is to isolate and characterize cellulose-degrading bacteria inhabiting these high pH serpentinizing ecosystems. A cellulose-based medium was adjusted to pH values of 8, 9, 10, and 12, and was inoculated with 4 different environmental samples obtained from various springs in the Philippines. Tubes were incubated at 35 °C in both anaerobic and aerobic conditions. After 3 weeks, growth was observed at all pH conditions, both aerobic and anaerobic, as evidenced by a distinct turbidity in the experimental tubes. DAPI microscopy was used to

verify presence of bacterial cells, and showed abundant cells of varying morphologies. DNA was extracted from cell cultures, followed by sequencing of the 16S gene in order to determine identities of individuals in culture. Continuing work in this study will aim to further characterize these cellulolytic strains, and gain an increased understanding of the biogeochemistry of these complex systems.

References:

[1] Schrenk M.O. and Brazelton W.J. (2013) *Rev Mineral Geochem*, 75, 575-606. [2] Gensel P.G. (2008) *Annu Rev Ecol Evol Syst.*, 39, 459-477.