

# Single-Cell Amplified Genomes from a Deep-Sea, Serpentinizing, Mud Volcano

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Deep-sea serpentinization occurs when low-silica ultramafic rocks rich in olivine and pyroxene interact with water, releasing life-fueling compounds such as ferrous iron, methane, and diatomic hydrogen [1]. This is one reason why oceanic serpentinization is proposed as an incubator for early life on Earth, and modern serpentinizing sites are early-Earth analogs [2]. These locales are also CO<sub>2</sub>-limited and hyper-alkaline, with pH at the limits of known biological habitability [1]. The Mariana forearc is an oceanic, active site of serpentinization within the Izu-Bonin-Mariana (IBM) convergent margin where the subducting slab releases fluids that expel at the seafloor forming massive alkaline serpentinite mud volcanoes [3]. These mud volcanoes have distinct chemistries and exist across a natural gradient of serpentinization-derived geochemical states [3]. Furthermore, they may host unique autotrophic communities that use serpentinization-derived compounds, such as CO, as a carbon and energy source. Microbial characterization of IBM mud volcanoes provides the opportunity to study an early-Earth analog and link microbial metabolism to subduction processes. To date, logistical sampling constraints and low biomass have impeded efforts to comprehensively characterize microbial life at IBM mud volcanoes. However, the generation of single-cell amplified genomes provides an opportunity to identify microbial members of this low-biomass system.

Here, we report single-cell amplified genomes (SAGs) generated from IBM mud volcano sediment samples collected during International Ocean Discovery Program Expedition 366. We classify multiple SAGs as members of the candidate phylum NPL-UPA2. Members of NPL-UPA2 have various features proposed in the last universal common ancestor and have been hypothesized to use CO for acetogenesis via the Wood-Ljungdahl pathway [4]. The SAGs in this study provide evidence that members of NPL-UPA2 reside within IBM mud volcanoes, and recruitment of recently published metatranscriptomes [5] to these SAGs is in progress to evaluate the contribution of SAGs to microbial community-level expression.

[1] Schrenk *et al.* (2013) *Reviews in Mineralogy and Geochemistry* 75(1), 575-606. [2] Russell *et al.* (2010) *Geobiology* 8(5), 355-371. [3] Fryer *et al.* (2017) *International Ocean Discovery Program Expedition 366 Preliminary Report*. [4] Suzuki *et al.* (2018) *Frontiers in Microbiology* 9, 3141. [5]