Hyperthermophile-Mineral Interactions and Correlating Mineral Transformations.

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Introduction: Dissimilatory iron reduction is the microbial respiration of Fe^{3+} to Fe^{2+} and is an important biogeochemical process in the anoxic subsurface [1], [2]. It may have been an important process on the early Earth, and could be a metabolism for life beyond Earth. Hyperthermophiles are organisms that grow optimally above 80°C, and represent life forms found early in life history. Our study sought to determine the biogenic minerals that form due to hyperthermophile-mineral interactions and the potential this has to be a biosignature.

Iron reduction in deep-sea hydrothermal vents has been largely overlooked due to the reduced nature of most minerals present and the insolubility of Fe^{3+} in circumneutral fluids. However, iron oxide minerals in the form of ferrihydrite [$Fe(OH)_3$] are common in mildly reducing hydrothermal systems due to seawater oxidation of iron sulfide minerals and could serve as an oxidizing agent for microbial respiration [3]. Indeed, hyperthermophilic iron reducers were ubiquitous in hydrothermal sulfide deposits collected from the Endeavour Segment hydrothermal vents [4].

Here, we examine the mineralogy of hyperthermophile reduction of ferrihydrite by two novel archaea, *Hyperthermus* sp. Ro04 and *Pyrodictium* sp. Su06 isolated from actively venting hydrothermal sulfide chimneys from the Endeavour Segment.

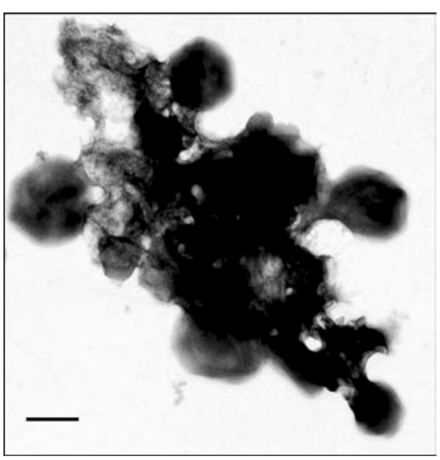

Fig. 1. Negative-stain transmission electron microscopy of *Hyperthermus hephaistosi* with attachment to iron oxide particles. The scale bars represent 700 nm.

We report their rates and constraints of Fe^{2+} production and the mineral end-products of their growth using using Mössbauer spectroscopy.

Characterization of growth: *Hyperthermus* sp. Ro04 and *Pyrodictium* sp. Su06 were grown anaerobically at 90°C and pH. 6.8 in Kashefi Marine Medium C [5] unless otherwise stated. The oxidizing agent was 100 mmol per liter of laboratory-synthesized ferrihydrite. The carbon and energy sources were peptides for Ro04 and H_2 and CO_2 for Su06. For each growth experiment, cell concentrations were determined at multiple time points using acridine orange stain and epifluorescence microscopy and Fe^{2+} concentrations were determined spectrometrically using the ferrozine assay.

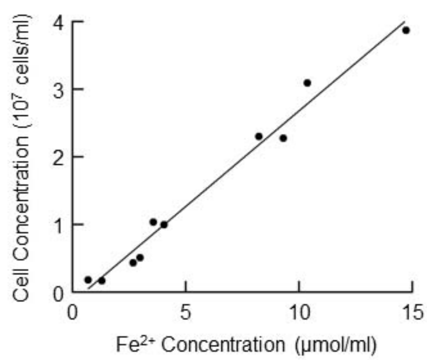

Fig. 2. Cell yield of *Hyperthermus hephaistosi* based on Fe^{2+} production (Y_{Fe2+}) calculated from a best-fit linear regression line through cell concentrations versus Fe^{2+} concentrations for one growth curve at 87°C.

Characterization of mineral products: Mössbauer spectra were determined for various iron oxide standards, abiotic controls, and biogenically reduced iron oxides from Ro04 and Su06. Pre- and post-growth iron oxides were dried and sealed in bottles in an anoxic chamber prior to analysis. Mössbauer spectra were acquired at Mount Holyoke College using 16 different temperatures ranging from 4 K to 295 K and calibrated relative to the spectrum of a 25 μm Fe foil. Both the Mexdist and Mexfield programs from the University of Ghent, which solve for the full Hamiltonian, were used to model the spectra. Errors on results for well-resolved components are usually ca. 0.02-0.04 mm/s for isomer shifts, quadrupole splitting and linewidths,

~0.1-0.3 Tesla for magnetic hyperfine fields, and 1-3% (absolute) for relative areas of distributions.

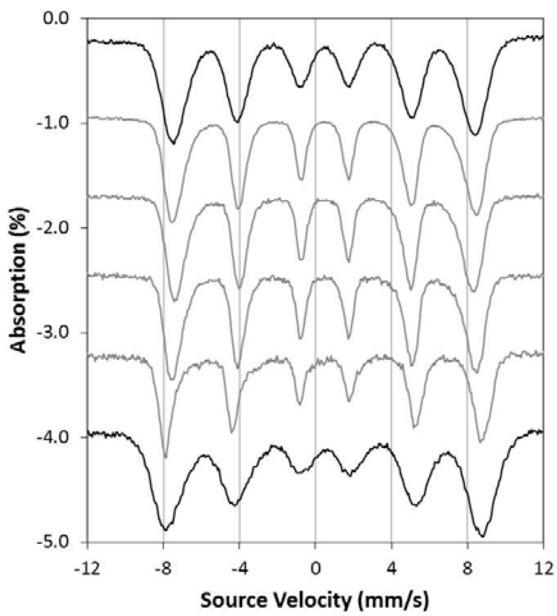

Fig. 4. Mössbauer spectra at 4 K of the iron oxide starting material (1), uninoculated growth medium before (2) and after (3) incubation at 90°C for 24 h, and the medium following growth of *Hyperthermus hephaistosi* to late logarithmic growth phase (4), along with nanophase ferrihydrite and magnetite standards. The reported IS and QS for the room temperature magnetite standard pertains only to the doublet.

Results and Discussion: A large variety of minerals are found in actively venting sulfides at hydrothermal vents. These minerals that have precipitated to form these sulfides have beginnings either from seawater or from hydrothermal fluid; they play a significant role in microbial composition especially in influencing the growth of microbes that can utilize these minerals. High-temperature-loving iron reducers, be they heterotrophs or autotrophs, are found ubiquitously and serve as a set of microbes that can take advantage of this milieu of minerals. This study provides a building block to understanding the intricate microbe-mineral interactions that must occur in order for life to use an insoluble electron acceptor.

Pure cultures were obtained from actively venting sulfide structures and the two novel hyperthermophilic iron reducers isolated from these samples. We have so far characterized growth rates of both organisms and correlated it with growth yield and Fe^{2+} production rates. Mineral transformations were also determined for both organisms and their mineral end products show differences in which minerals are produced by biogenic iron reduction. Both growth data and mineral data can help us determine possible location of these organisms and could also suggest the presence of these organisms in terrestrial analogs if potential biosignatures are found.

References:

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