

## B21D-06 1145h INVITED

**Anaerobic Oxidation of Methane Mediated by Microbial Consortia**

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Stable isotope signatures, radiotracer and modeling techniques have established that most of the methane rising from cold seeps, mud volcanoes and gas hydrates is oxidized microbially under anoxic conditions. However, details of the related biochemical mechanisms and organisms are still largely unknown, although the anaerobic oxidation of methane (AOM) is the major biological sink of methane in marine sediments and thus crucial in balancing the emission of this important greenhouse gas into the atmosphere. The isotopic and genetic signature of the microbial biomass in methane-saturated seep sediments shows that AOM is mediated by different microbial consortia which generally include archaea and sulfate-reducing bacteria. Fluorescence in situ hybridization revealed that both archaea and SRB grow together in symbiotic association. Yet, it cannot be ruled out that single archaea might exist which are capable of anaerobic oxidation of methane. Among the archaea from gassy sediments, rRNA probes target specifically the ANME-2 group, belonging to the Methanosarcinales, and the ANME-1 group. A new cluster of delta-proteobacterial sulfate reducers that is closely related to the *Desulfosarcina-Desulfococcus* group is also found in most methane rich environments. The consortia are embedded in an organic matrix probably serving as a nucleus for calcification. Field and laboratory data indicate that the process of AOM can support significant cell growth and activity, despite the very low energy yield predicted by thermodynamic models.

**B22A MC: Hall D Tuesday 1330h****Biological Mineralization: Proxies and Processes (joint with OS, PP, MR)**

**Presiding:** P M Dove, Virginia

Polytechnic Institute and State

University; J J DeYoreo, Lawrence

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**B22A-0124 1330h POSTER****The Mineralogy and Microstructure of Sedimentary Zinc Sulfides Formed by Bacterial Sulfate Reduction.**

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Bacterial sulfate reduction (BSR) is considered to be the predominant mechanism for low-temperature conversion of sulfate to sulfide [1] and is inferred to have existed since the early Proterozoic [2, 3]. Because BSR leads to precipitation of abundant metal sulfide minerals, some ancient, low-temperature sedimentary ore deposits are now hypothesized to have biogenic origins [4]. We have studied zinc sulfide minerals produced by sulfate-reducing bacteria (SRB) living in anoxic, 8 °C waters of a flooded mine near Tenynson, Wisconsin [5]. Our objectives were to characterize the morphology, mineralogy, and microstructure of the biominerals and to look for potential biosignatures. Scanning electron microscope images from cryofixed, freeze-fractured samples and transmission electron microscope (TEM) images from ultramicrotomed samples

show a close association between cells and spherical aggregates of ZnS. However, SRB cells are generally not coated by ZnS, implying that the particles form and aggregate in solution after sulfide is expelled from the cell. High-resolution TEM images reveal that the few-micron diameter spheres are comprised of about a billion ZnS particles that are typically 1.5-5 nm in diameter. More coarsely crystalline regions appear to have grown via oriented aggregation of smaller nanoparticles. In some cases, orientation gives rise to twinning on {111} sphalerite. ZnS particles are primarily sphalerite, but domains of wurtzite are not uncommon. Even some of the smallest particles have periodic structure and well-defined morphologies. Reasons for the formation of wurtzite remain unclear, but may be related to the sulfide concentration during aggregation of multinuclear clusters [6] or size-dependent phase stability. In addition, the ZnS spheres are not of uniform density throughout, but contain concentric zones separated from each other by 5-7 nm-wide (average) regions of low particle density. The number of zones per sphere is variable, as is the width of each concentric layer. The existence of these zones implies periodic fluctuations of some form, possibly correlated to nutrient availability, changes in redox conditions within the biofilm, or both. Qualitatively, some spheres located within the same region of the biofilm display similar concentric patterns, indicating that fluctuations are coordinated over micron-scale regions. These results reveal a number of mineralogical and microstructural characteristics of biogenic ZnS that constitute potential biosignatures. These characteristics may be present and recognizable in ancient sedimentary ZnS deposits and would support a nanocrystalline, low-temperature, biogenic origin.

References: [1] Trudinger et al. (1985) *Can J. Earth Sci.* 22, 1910-1918; [2] Ohmoto et al. (1993) *Science* 262, 555-557; [3] Wagner et al. (1998) *J. Bacteriol.* 180, 2975-2982; [4] Anderson et al. (2001) *Econ.Geol.* 96, 885-890; [5] Labrenz et al. (2000) *Science* 290, 1744-1747; [6] Luther et al. (1999) *Geochim. Cosmochim. Acta* 63, 3159-3169.

**B22A-0125 1330h POSTER****Microbes make average 2 nanometer diameter crystalline UO<sub>2</sub> particles.**

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It is well known that phylogenetically diverse groups of microorganisms are capable of catalyzing the reduction of highly soluble U(VI) to highly insoluble U(IV), which rapidly precipitates as uraninite (UO<sub>2</sub>). Because biological uraninite is highly insoluble, microbial uranyl reduction is being intensively studied as the basis for a cost-effective in-situ bioremediation strategy. Previous studies have described UO<sub>2</sub> biomineralization products as amorphous or poorly crystalline. The objective of this study is to characterize the nanocrystalline uraninite in detail in order to determine the particle size, crystallinity, and size-related structural characteristics, and to examine the implications of these for reoxidation and transport.

In this study, we obtained U-contaminated sediment and water from an inactive U mine and incubated them anaerobically with nutrients to stimulate reductive precipitation of UO<sub>2</sub> by indigenous anaerobic bacteria, mainly Gram-positive spore-forming *Desulfosporosinus* and *Clostridium* spp. as revealed by RNA-based phylogenetic analysis. *Desulfosporosinus* sp. was isolated from the sediment and UO<sub>2</sub> was precipitated by this isolate from a simple solution that contains only U and electron donors. We characterized UO<sub>2</sub> formed in both of the experiments by high resolution-TEM (HRTEM) and X-ray absorption fine structure analysis (XAFS).

The results from HRTEM showed that both the pure and the mixed cultures of microorganisms precipitated around 1.5 - 3 nm crystalline UO<sub>2</sub> particles. Some particles as small as around 1 nm could be imaged. Rare particles around 10 nm in diameter were also present. Particles adhere to cells and form colloidal aggregates with low fractal dimension. In some cases, coarsening by oriented attachment on {111} is evident. Our preliminary results from XAFS for the incubated U-contaminated sample also indicated an average diameter of UO<sub>2</sub> of 2 nm. In nanoparticles, the U-U distance obtained by XAFS was 0.373 nm, 0.012 nm smaller than found in the bulk structure of UO<sub>2</sub> (0.385 nm). This indicates contraction within the nanoparticles due to tensile surface stress. Microbially formed UO<sub>2</sub> is highly reactive, thus will be oxidized quickly as redox conditions change.

Our findings support a growing number of studies that indicate that biominerals formed as the result of enzyme-mediated redox reactions are nanoparticulate.

Preliminary results suggest that these particles will be readily transported through sandy aquifers, especially when conditions prevent high degrees of flocculation. Thus, despite its low (but size-dependent) solubility, UO<sub>2</sub> nanoparticle transport may exert a fundamental control on mobility of U in contaminated environments.

**B22A-0126 1330h POSTER****The Effect of Bacterial Surfaces on Silica Precipitation**

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Bacterial silicification is an important geological process in modern geothermal environments (e.g., New Zealand, Iceland, Japan). The precipitation of silica onto bacterial surfaces can affect microbial fossilization, chemical sediment formation, the porosity and permeability of crustal rocks, and silica transport in geothermal hot springs. Previous studies have suggested that active deposition of silica onto bacterial cells begins with the precipitation of heterogeneously nucleated aggregates of amorphous silica. However, the effect of bacteria on silica precipitation is poorly understood, and it is unclear if bacterial surfaces enhance the kinetics of silica precipitation or if the bacteria act as passive precipitation surfaces.

In this study, we performed silica precipitation experiments with the filamentous cyanobacteria *Calothrix* sp. (strain KC97) to elucidate the rates and mechanisms of silicate biomineralization. Batch experiments were conducted as a function of time, Si saturation states, temperature, pH and Fe concentrations. Experiments at both undersaturated and supersaturated conditions indicate that Si-bacteria interactions are weak, and that minimal bacterial silica sorption/precipitation occurs. In supersaturated solutions, abiotic polymerization rates are rapid and at the times scales of our experiments (1-300 hours) the presence of bacteria does not enhance silica nucleation or monomeric silica polymerization. However, the presence of Fe-coated bacteria significantly increases silica sorption/precipitation rates, and the extent of Si sorption/precipitation increases with increasing Fe concentrations. Fe precipitation experiments performed without Si indicate that iron precipitation onto bacterial surfaces occurs very rapidly and significantly faster than abiotic controls. The experimental data suggest that in the presence of Fe, bacterial silicification occurs as a two step process: 1) Fe precipitates onto bacterial cells via heterogeneous surface nucleation, followed by 2) Si sorption/precipitation onto the Fe-coated bacterial surface. Therefore, Fe may play an important role in the silicification of microbial cells in geothermal environments.

**B22A-0127 1330h POSTER****Manganese Oxide Biomineralization by Spores of the Marine Bacillus sp. Strain SG-1**

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Biogenic Mn oxides are ubiquitous in natural waters, have high sorptive capacities for metal ions, and oxidize organic and inorganic substances such as aromatic hydrocarbons, Cr(III), and hydrogen sulfide. In this fashion, Mn(II)-oxidizing bacteria impact the biogeochemical cycling of essential nutrients and toxic trace constituents of natural waters. In spite of their importance, the molecular mechanisms, intermediates, and products of Mn oxide biomineralization are poorly understood. Similarly, the relationship between biotic

and abiotic Mn oxidation mechanisms is not well documented.

We have studied Mn oxide biomineralization by spores of the marine *Bacillus* sp. strain SG-1 as functions of reaction time (10 min to 77 d), Mn(II) concentration (0.01 to 1 mM), major ion composition (50 mM NaCl to sea water), O<sub>2</sub> partial pressure, and temperature. SG-1 spores are an ideal subject because they are dormant, Mn-oxidation is not inactivated by the x-rays utilized, and they previously have been extensively studied. Reaction products and Mn oxidation state evolution were directly observed in order to infer mechanisms and phase dominance. To obtain this information, a combination of Mn(II) uptake measurements, K-edge x-ray absorption spectroscopy (XAS), L-edge scanning transmission x-ray microscopy (STXM, 60 nm nominal spot size), and synchrotron-based x-ray diffraction measurements were performed. All samples were measured under fully hydrated conditions to prevent dehydration of reaction products. This set of techniques provides chemical and structural information on Mn in amorphous and crystalline states in the samples.

Mn oxide biomineralization products were sensitive to [Mn(II)]. At 0.01 mM [Mn(II)], biogenic Mn oxides were found to contain highly oxidized Mn (80-85% Mn(IV)) as observed after 48 hr. reaction. The dominant phase is identified as an amorphous Mn(IV) oxide similar to d-MnO<sub>2</sub>. K-edge XAS measurements suggest this phase forms within minutes of reaction onset and dominates after 6 hr. reaction, without detection of Mn(III). In contrast, L-edge STXM suggests that Mn(III) is important throughout the 6 - 48 hr interval and Mn(IV) is a minor component. At [Mn(II)] = 1 mM, crystalline b-MnOOH was observed as a dominant abiotic reaction product, likely formed on biogenic am. MnO<sub>2</sub> surfaces. We will discuss important aspects of K- and L-edge spectroscopy and their application on microscopic scales and demonstrate how these aspects account for each set of observations. Implications for reaction mechanisms will be presented.

#### B22A-0128 1330h POSTER

##### Environmental Controls of Biological Manganese Oxidation

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Biological catalysis of manganese oxidation represents an important contribution to global manganese cycling; biological oxidation rates are several orders of magnitude higher than those of abiotic processes. Despite recent genetics advances, ongoing behavioral studies, and a large pool of knowledge regarding manganese chemistry, the links between biology and environmental chemistry remain unresolved. We have performed experiments on batch cultures of *Leptothrix discophora* SS-1 to explore the physiology of biological manganese oxidation. We have further conducted spectroscopic and microscopic studies of the mechanism as manganese proceeds from the soluble Mn<sup>2+</sup> species to the insoluble Mn(III) and Mn(IV) phases. These investigations suggest roles for aqueous chemistry, mineralogy, and microbial physiology in controlling manganese fluxes in metal-rich environments.

#### B22A-0129 1330h POSTER

##### Diurnal Changes in Microstructure and Microscale Chemistry of Reef Coral Skeleton

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Corals with symbiotic zooxanthellae exhibit large diurnal fluctuations in calcification rate associated with the photosynthetic activity of their symbionts. An obvious way in which photosynthesis can enhance calcification is by removing free carbon dioxide from the medium in which crystal growth occurs, increasing alkalinity and the availability of carbonate ions. We investigated the effects of photosynthesis on the ultrastructure and elemental chemistry of aragonite crystals which constitute the growing skeleton. Sclerodermites are the basic building blocks of the skeleton, each consisting of a center of calcification and its associated fasciculi. Each night, a new center of calcification is

deposited on the tips of pre-existing skeletal elements. These crystals are submicron in size and equant in shape, indicative of slow precipitation in a high CO<sub>2</sub> environment. During the day a dense array of long, needle-shaped crystals grow out from the centers in organized bundles (the fasciculi) and these constitute the bulk volume of the skeleton. Their morphology is characteristic of crystals which grow rapidly in a low CO<sub>2</sub> environment. We measured changes in strontium-calcium content (Sr/Ca) of the sclerodermite over the diurnal cycle using Secondary Ion Mass Spectrometry (SIMS), an in-situ microbeam measurement technique. We observe a linear decrease in skeletal Sr/Ca as the sclerodermite grows to fill the extracellular calcifying space. Sr/Ca in crystals accreted at night is close to equilibrium values, decreasing along the length of the needle-shaped crystals as these grow during the day. Our observations indicate that the elemental chemistry of zooxanthellate coral skeleton is strongly influenced by kinetic processes which override equilibrium processes when crystal growth rates are high. Clearly, photosynthetic activity of algal symbionts in coral tissue plays a role in determining both the microstructure and chemistry of coral skeleton over the course of a single day. Our results have important implications for the use of coral Sr/Ca as a paleotemperature proxy.

#### B22A-0130 1330h POSTER

##### Investigation of Strontium Incorporation into Biotically and Abiotically Precipitated-Calcium Calcite Using Secondary Ion Mass Spectrometry

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Radionuclide and metal contaminants are present in the vadose zone and groundwater throughout the U.S. Department of Energy (DOE) complex. A possible approach to their remediation is in situ immobilization by co-precipitation of these elements in authigenic calcite and calcite overgrowths. Microorganisms are known to facilitate the precipitation of calcite; hence the stimulation of biogenic calcite production may offer a means to accelerate co-precipitation of contaminant metals. Strontium is well-known to substitute for Ca in calcium carbonate minerals, and consequently, the uranium fission product <sup>90</sup>Sr is a prime candidate for this type of remediation approach.

In order to predict the extent and stability of Sr incorporation into calcite precipitated under this bioremediation strategy, it is necessary to understand how much Sr is being incorporated. In these studies, secondary ion mass spectrometry (SIMS) was utilized to characterize the surface chemistry of carbonates generated by bacterial activity in synthetic groundwater containing Ca and Sr. SIMS with sputter depth profiling allows the determination of changes in Sr to Ca ratios with depth in particulate carbonate samples. The sputter depth profiling results can be compared with analysis of the bulk composition by inductively coupled plasma atomic emission spectroscopy (ICP-AES). Results of analyses on carbonates generated by *B. pasteurii* in synthetic groundwater with initial Ca and Sr concentrations of 80 ppm and 10 ppm, respectively, showed that SIMS could successfully measure ion ratios on the surface and within these particles. ICP-AES data indicated a bulk Sr:Ca ratio of 0.11, and sputtering SIMS data approached this value with increasing depth into the particle. The Sr:Ca ratio however, contrary to what would be expected from precipitation under batch conditions, was lower at the surface of the particles (ca. 0.05) and increased with depth. One possible reason for this phenomenon is re-equilibration with solution conditions after initial fast precipitation induced by the microorganisms. In this presentation, we will evaluate the utility of using SIMS for verifying microbially accelerated production of carbonates for remediation by comparing biotically generated carbonates to carbonates precipitated under abiotic conditions.

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#### B22A-0131 1330h POSTER

##### Biological control on Sr partitioning during calcification of *Emiliania huxleyi*

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The concentration of trace elements (e.g. Sr, Cd, Ba and Zn) in marine biogenic calcites has the potential to act as a tool to probe past ocean conditions and

related climate change. The concentration of a trace element such as Sr in the calcite depends on two factors: the oceanic concentration of Sr, and the partitioning of Sr between seawater and the biogenic calcite. We challenge the view that the partitioning of trace elements occurs according to predictions for inorganic calcite precipitation and propose that the chemistry of the biogenic calcite is instead controlled by biological discrimination during the calcification process. As an example, we investigate the partitioning of Sr and Ca during calcification with culture experiments of the coccolithophore *Emiliania huxleyi*. As the calcite liths are constructed in an intracellular vesicle during calcification, we propose that the biological discrimination between the similar Sr<sup>2+</sup> and Ca<sup>2+</sup> ions by Ca<sup>2+</sup>-selective channels and pumps, or the organic template, is the dominant control on the chemistry of the calcite. An understanding of environmental controls on the biological discrimination between Ca and trace elements during calcification is vital for using these paleoceanographic tools to record past climates.

#### B22A-0132 1330h POSTER

##### Understanding the Molecular-Scale Processes of Biomineralization: Resolving the Physical Mechanism by which Mg<sup>2+</sup> Modifies Calcite Morphology

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Magnesium is the principal modifier of calcite morphology in natural waters and is a key impurity in carbonate biominerals. However, the molecular-scale mechanism by which magnesium determines calcite morphology has remained controversial due to a lack of direct experimental insight. Here we use in situ molecular-scale observations of step dynamics and growth hillock morphology to directly resolve the role of magnesium in governing calcite surface morphologies. We show that the interaction of magnesium with monomolecular steps depends upon the molecular-scale structure of the step-edge. These step-specific interactions are especially evident at low Mg/Ca ratios in solution, where steps with acute step-edge geometries are observed to be rough while obtuse steps remain smooth. Higher Mg/Ca solution ratios cause both step-types to become rough and the growth spiral to assume a more isotropic form. In addition, new step-directions are generated parallel to the c-glide plane. These new step directions are shown to be the result of lattice mismatch at the intersection of nonequivalent step-types, resulting from differential magnesium incorporation into those steps. The resultant strain at the boundary of the nonequivalent step-types leads to decreased growth rates and the development of new step directions parallel to the c-axis. These observations offer a plausible molecular-scale mechanism for macroscopic calcite morphologies that are elongated along the c-axis. Further, this mechanism extends our understanding of the role that crystal structure plays in determining mineral morphology by demonstrating that surface morphologies are controlled by differences in the molecular-scale structure of otherwise crystallographically equivalent steps. Finally, this novel mechanism of calcite habit modification by magnesium provides needed insight into the biomineralization strategies required to produce the exquisite crystal morphologies present in biomineralizing systems.

#### B22A-0133 1330h POSTER

##### An Atomic Scale Look at the Thermodynamics and Kinetics of Mineralization

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A common theme in biomineral growth is that biological systems use peptides and proteins to modify nucleation and growth of the inorganic phase. Developing a detailed physical picture of this process at the atomic-scale mechanisms naturally begins with an understanding of crystallization in the organic-free systems. Over the past eight years, atomic force microscopy has been used to investigate the growth of a wide variety of crystals from solution including many relevant to mineralization in biological systems such as calcium carbonate and calcium phosphate. The ability to observe growth in situ at molecular length scales using controlled solution compositions and conditions has led to significant advances in our understanding of crystal growth both in pure solutions and those containing organic additives that modify the growth morphology and kinetics. In many cases, the observed behavior has diverged significantly from that expected based on accepted atomistic models of growth. Here we use results of in situ AFM studies on calcite and brushite to illustrate aspects of our current understanding of mineralization that appear to be on a sound footing, and to highlight those areas where fundamental questions still remain unanswered. We examine three aspects of growth: step generation, step dynamics, and step kinetics. We find that analysis of the supersaturation dependence of step generation and kinetics call into question rough step models of growth that assume the applicability of the Gibbs-Thomson effect. We show that non-linear dependencies of step kinetics on supersaturation are an immediate consequence of a smooth step model of growth. We use measurements of step edge fluctuations and terrace width distributions to determine the extent of step-step interactions and the pathways of mass transport at step edges. The significance of these results for understanding the role of organic modifiers will be discussed.

#### B22A-0134 1330h POSTER

##### Using Microbial Anoxic Experiments to Study Oxygen Isotope Fractionation in Dolomite.

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Modern microbial Ca-dolomite precipitating in a shallow hypersaline coastal lagoon (Lagoa Vermelha, Brazil) furnished information about the mechanisms and conditions promoting the mineral formation. Bacterial culture experiments, were conducted using a synthetic liquid medium which simulated the chemical conditions of this natural environment. The medium has two time seawater salinity and formate as a carbon source. Dolomite was produced during an incubation time of 30 days. Although, microbes have optima growth temperatures, it was necessary to conduct experiments at different non-optimal temperatures in order to examine the oxygen-isotope equilibrium between dolomite and water.

So far, dolomite has been precipitated at 25, 30, 35, 40 and 45 °C. In order to grow cells under non-optimal conditions, the experiments had to be adapted by increasing the cell density because of the low cell growth rates at low temperatures. Also, additional substrate was added to the media during the incubation to sustain for sulfate reduction during the entire duration of the experiment. As a consequence, changes in the media chemistry must be taken into consideration. Our preliminary results show that at 25 and 30 °C, a mixture of high Mg-calcite and Ca-dolomite formed, whereas at 40 and 45 °C pure stoichiometric dolomite was obtained. The  $\delta^{18}\text{O}$  value of dolomite precipitating at 30 and 40 °C differs by  $1.8\text{‰}$ .

#### B22A-0135 1330h POSTER

##### Ion microprobe U-Pb dating and REE abundance of biogenic apatite

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If the direct U-Pb dating of a fossil itself is possible, the method could have great impact on stratigraphic studies in establishing the absolute chronology

of sedimentary sequences. Micro fossil ?conodont? are candidates for this purpose since they consist of apatite ( $\text{Ca}_2(\text{PO}_4)_3(\text{F,Cl,OH})$ ), which would uptake U, Th and Pb after sedimentation no longer than a few million years and is supposed to remain closed to U and Pb under relatively low effective closure temperature. We report here results of direct ion microprobe U-Th-Pb dating of two conodonts; Trichognathus from Kinderhookian stage of Mississippian sedimentary sequence from Illinois Basin region in North America and Panderodus from a Llandoveryan sedimentary sequence on Langkawi Island, northern Malaysia. Secondary purpose of the study is to indicate in situ analysis of all REE on the same spots of U-Pb measurements. Samples were cast into epoxy resin discs with a few grains of standard apatite, PRAP, derived from an alkaline rock of Prairie Lake circular complex in the Canadian Shield and polished until they were exposed through their mid-sections. U, Th and REE abundances, and Pb isotopic compositions were measured by using SHRIMP installed at Hiroshima University. Thirteen spots on Trichognathus yield a  $^{238}\text{U}/^{206}\text{Pb}$  isochron age of  $323\pm 36$  Ma, which is consistent with the depositional and early diagenetic ages. Fifteen spots on Panderodus give  $^{232}\text{Th}/^{208}\text{Pb}$  isochron age of  $429\pm 50$  Ma, which is again comparable to an early Silurian. Shale-normalized REE of Trichognathus shows a broadly flat pattern from light to middle REE and a decrease from middle to heavy REE with negative anomalies of Ce and Eu. In contrast Panderodus indicates a concave-shape pattern with middle REE enrichment. These characteristics are probably due to a different formation environment as suggested by other workers.

#### B22A-0136 1330h POSTER

##### Incorporation of Trace Elements in Ancient and Modern Human Bone: An X-Ray Absorption Spectroscopy Study

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X-ray absorption spectroscopy (XAS) affords the opportunity to probe the atomic environment of trace elements in human bone. We are using XAS to investigate the mode(s) of incorporation of Sr, Zn, Pb, and Ba in both modern and ancient (and thus possibly altered) human and animal bone. Because burial and diagenesis may add trace elements to bone, we performed XAS analysis on samples of pristine contemporary and ancient, buried human and animal bone. We assume that deposition of these elements during burial occurs by processes distinct from those *in vivo*, and this will be reflected in their atomic environments.

Archaeologists measure strontium in human and animal bone as a guide to diet. Carnivores show lower Sr/Ca ratios than their herbivore prey due to discrimination against Sr relative to Ca up the food chain. In an initial sample suite no difference was observed between modern and buried bone. Analysis of additional buried samples, using a more sensitive detector, revealed significant differences in the distance to the second and third neighbors of the Sr in some of the buried samples. Distances to the first neighbor, oxygen, were similar in all samples.

Zinc is also used in paleo-diet studies. Initial x-ray absorption spectroscopy of a limited suite of bones did not reveal any differences between modern and buried samples. This may reflect the limited number of samples examined or the low levels of Zn in typical aqueous solutions in soils. Signals from barium and lead were too low to record useful XAS spectra. Additional samples will be studied for Zn, Ba, and Pb.

We conducted our XAS experiments on beam lines 4-1 and 4-3 at the Stanford Synchrotron Radiation Laboratory. Data were collected in the fluorescence mode, using a Lytle detector and appropriate filter, and a solid state, 13-element Ge-detector.

#### B22A-0137 1330h POSTER

##### Multiscale Topographical Analysis of Biogeochemically Reduced Hematite Surfaces

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Establishing the mechanisms and magnitudes of nano-mesoscale influences on interfacial chemical reactivity requires a multiscale description of the structure of the interfacial region. The identification of scaling relationships characterizing mineral surface structure in low-temperature environments is a first step in the construction structure-activity relationships that are potentially applicable over multiple length scales. Using wavelet image processing techniques and scaling relationships such as the evaluation of Hurst exponents and fractal dimension, we systematize and quantify mineral surface topography of a sample of hematite undergoing biochemically induced reductive dissolution. Image mosaicking methods commonly applied in remote sensing and medical imaging contexts are applied to AFM images to obtain large scale images for the evaluation of scaling exponents. Gaussian wavelet methods are used to enhance and quantify structural features associated with the biogeochemically reduced surfaces.

#### B22A-0138 1330h POSTER

##### Effects of Natural Surfactants and Analogs on Siderophore Controlled Weathering Reactions

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It has been shown that a wide variety of microorganisms produce and exude surfactants with a range of chemical structures and properties. Adsorption of surfactants to mineral surfaces has been investigated previously. The modification of the mineral surface chemistry by adsorbed surfactants plays an important role in bacterial attachment. The purpose of this research was to elucidate the effect of bacterial surface active compounds and synthetic analogs on important biogeochemical processes such as mineral weathering and bacterial iron acquisition. In this context we studied the influence of surfactants on goethite ( $\alpha\text{-FeOOH}$ ) dissolution kinetics in the presence of the bacterial siderophore desferrioxamine-B (DFO-B). Siderophores are iron specific low molecular weight organic ligands that are exuded by microorganisms under iron deficient conditions.

We investigated the effect of surface active compounds including rhamnolipid (a bacterial surfactant), fatty acids, and sodium dodecyl sulfate (a synthetic surfactant) on siderophore mediated iron oxide dissolution. Dissolution and adsorption experiments were conducted at pH=6. At this pH, all studied surfactants are anionic.

At constant dissolved siderophore concentrations, we observed increasing goethite dissolution rates with increasing concentrations of bacterial and synthetic surfactants. In control experiments, we verified that the surfactants alone have no effect on iron oxide dissolution. We also demonstrated that increasing surfactant concentrations are associated with an increase in adsorbed DFO-B concentrations. Comparing adsorbed DFO-B concentrations as a function of surfactant concentrations with dissolution rates, we found a linear correlation.

A linear correlation between adsorbed ligand concentrations and dissolution rates is an indicator for a ligand controlled dissolution mechanism. Thus it appears that surfactants influence DFO-B mediated dissolution indirectly by increasing the affinity of the siderophore for adsorption at the goethite surface. This is caused, at least in part, by the influence of the anionic surfactants on the mineral surface charge. We observed charge reversal of the initially positively charged goethite surface with increasing surfactant concentrations. This removes electrostatic repulsion between the positively charged ligand and the (initially) equally charged surface. However, an additional hydrophobic effect can not be ruled out at this time and needs to be investigated in the future.

#### B22A-0139 1330h POSTER

##### Biogeochemistry and Spatial Distribution of the Microbial-Mineral Interface Using I<sup>2</sup>LD-FTMS

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Previous studies indicate that biogeochemistry can vary within individual mineral specimens in contact with microorganisms. These same studies have shown that microcosms containing a mixture of minerals simulating a heterogeneous geologic matrix do not yield the same results as the naturally occurring rock. Therefore, it is of utmost importance to develop analytical tools that can provide spatially correlative biogeochemical data of the microbial-mineral interface within naturally occurring geologic matrices. Imaging internal laser desorption Fourier transform mass spectrometry (<sup>12</sup>LD-FTMS) can provide elemental and molecular information of the microbial-mineral interface at a spatial resolution limited only by the optical diffraction limit of the final focusing lens (down to 2 μm). Additionally, the <sup>12</sup>LD-FTMS used in this study has exceptional reproducibility, which can provide successive mapping sequences for depth-profiling studies. Basalt core samples, taken from the Snake River Plain Aquifer in southeastern Idaho, were mapped prior to, and after, exposure to a bacterial culture. The bacteria-basalt interface spectra were collected using the <sup>12</sup>LD-FTMS at the INEEL. Mass spectra were recorded over a mass-to-charge range of 30-2500 Da with an average peak resolution of 15,000 using 10 μm spots. Two-dimensional maps were constructed depicting the spatial distribution of the minerals within the basalt as well as the spatial distribution of the bacteria on the basalt surface. This represents the first reported application of <sup>12</sup>LD-FTMS in the field of biogeochemistry.

**B22A-0140 1330h POSTER**

**Fractionation of Natural Organic Matter Upon Adsorption to the Bacterium, *Bacillus subtilis***

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High pressure size exclusion chromatography (HPSEC) was used to measure changes in molecular weight distribution and average molecular weight upon adsorption of fulvic acid onto *Bacillus subtilis* at pH 3-7. The FA was an XAD-8 extract from a stream in the New Jersey Pine Barrens (USA), and had a weight average molecular weight of 1890 Da.

Adsorption of aqueous FA onto *B. subtilis* was relatively fast, with steady state attained within 2 hours. An adsorption isotherm at pH 4.5 revealed a strong affinity of FA for the *B. subtilis* surface. The maximum adsorption capacity of a 20g bacteria/L suspension was greater than 9 mg C/L of FA at pH 4.5.

Adsorption of FA onto *B. subtilis* was strongly pH dependent, increasing markedly with decreasing pH over the pH range 3-7. Comparison of HPSEC analysis of control (FA not reacted with bacteria) versus reacted samples showed that in all experiments, the weight average molecular weight (Mw) of FA remaining in solution decreased by several hundred Da. The observed decrease in solution Mw upon adsorption indicated that the higher molecular weight FA components adsorbed preferentially to the bacterial surfaces, at all studied pH values (3-7). Additionally, there was a low molecular weight FA fraction that did not adsorb, even at low pH. Our results suggest that hydrophobic interactions may be important for FA sorption to *B. subtilis* and that low molecular weight, more hydrophilic components may thus be less likely to adsorb than higher molecular weight, more hydrophobic components.

**B22A-0141 1330h POSTER**

**Mineralization of Iron Oxyhydroxides in the Presence and in the Absence of Bacterial Cells**

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Because of their small size, iron oxides have a large surface area per unit weight ratio and are believed to play an important role as an adsorbing phase in lake sediments for various molecules, including potentially dangerous ones like heavy metals. They have been observed to form in close association with bacterial cells, by oxidation of ferrous ions. It is thus important to determine whether the presence of the bacterial cells can affect the mineralogy and the mesoscopic structure of the Fe-oxides particles, as well as their reactivity towards heavy metals.

We synthesized in the lab nanoparticles of Fe-oxides by oxidation of ferrous ions. This was done in the presence and in the absence of various bacterial strains (*Escherichia coli*, *Bacillus subtilis*, *Pseudomonas Aeruginosa* and *Bacillus licheniformis*) and of inorganic ligands (sulfate, phosphate, silicate). The Fe-oxides particles were then observed by TEM on thin sections and on whole mounts. The chemical composition was estimated by wet chemistry and by EDS. The mineralogy was determined by XRD, SAED and EXAFS. Surface area was investigated by BET. And adsorption of cadmium was also measured at various pH.

We observed that the size and the morphology of the particles as well as their mesoscopic spatial organization can be affected by the presence of the cells, whereas the mineralogy is controlled by the chemistry of the solution. The adsorption isotherms of cadmium on the various Fe-oxides will be discussed at the light of these observations.

**B22A-0142 1330h POSTER**

**Enhanced Dissolution of Iron Oxides by Biogenic H<sub>2</sub>S Produced by a Sulfate-Reducing Bacterium**

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In natural environments, reductive dissolution is by far the most important dissolution mechanism for iron oxides, either biotically or abiotically. Enzymatic reduction of iron oxides by sulfate-reducing bacteria (SRB) has been reported (Lovley et al., 1993; Tebo and Obraztsova, 1998). The goal of this study was to determine the extent to which SRB can enhance iron reduction through production of biogenic H<sub>2</sub>S. Biologic experiments were performed using a *Desulfovibrio desulfuricans* G-20. Abiotic reduction of iron oxides was performed using commercial Na<sub>2</sub>S. When iron oxides were used as the sole electron acceptor, which included hematite, goethite, ferrihydrite, and magnetite, less than 10% of iron was reduced by G-20 after 240 hrs. When iron oxides and sulfate were both used as the electron acceptors, reduction of iron by G-20 ranged from 60% for hematite to 97% for magnetite during the same period of time. In the abiotic control, less than 5% of hematite and goethite but great than 45% of ferrihydrite and magnetite were reduced after 120 hrs. These results indicate that poorly crystalline iron oxides (such as ferrihydrite) or the partially reduced iron oxide magnetite is most susceptible for abiotic reduction by S<sup>2-</sup>. The crystalline goethite and hematite are least susceptible for abiotic reduction by S<sup>2-</sup>, but their reduction can be significantly enhanced by biogenic H<sub>2</sub>S through bacterial mediation. These results suggest that iron reduction in sulfidic environments is not purely an abiotic chemical process but may be mediated by the sulfate-reducing bacteria, particularly for diagenesis of goethite and hematite.

URL: <http://web.missouri.edu/~geosccc>

**B22A-0143 1330h POSTER**

**Microbially-Induced Reduction of Iron Oxides: Impact of Flow on Secondary Mineral Phase Products**

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Despite the acknowledged importance of ferric oxides in controlling contaminant behavior, there is limited understanding of the products and processes during reductive dissolution of these mineral phases under dynamic flow conditions. Factors potentially controlling the fate of iron during reduction include dissolved Fe and electron donor concentrations; therefore, the nature and reactivity of these products are expected to differ when a solute transport component is introduced. We have conducted column experiments using a common, iron-reducing bacterium (*Shewanella putrefaciens*, strain CN32) and 2-line ferrihydrite-coated silica within an artificial groundwater matrix. Changes in pore water chemistry were monitored and solids were characterized as a function of time and flow path (2-cm intervals) using a variety of methods including x-ray absorption fine structure (XAFS) and high-resolution transmission electron microscopy (TEM). Microbial Fe(III) reduction results in the conversion of ferrihydrite to goethite and magnetite. Sequestration of Fe(III) within these secondary phases results in decreased Fe(II) bioavailability and subsequent decreased rates of microbial reduction. Magnetite likely forms via topotactic conversion of ferrihydrite and is correspondingly associated with the ferrihydrite surface, while goethite appears to form via dissolution and reprecipitation and is concentrated on the cell envelope. Changes in flow velocity result in dramatic differences in secondary mineral phase accumulation suggesting that linking hydraulic conditions with biogeochemical processes is an important step to predict the fate of Fe and associated trace metals during reductive dissolution of iron oxides.

**B22A-0144 1330h POSTER**

**The Role of Biogeochemical Dynamics in the Alteration of Uranium Solid Phases Under Oxidic Conditions.**

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Microbial reduction of uranium has been shown to lower groundwater concentrations of uranium in anoxic systems, but such biological alterations must be considered temporary unless long-term anoxia can be guaranteed. Under oxidic conditions, the more soluble higher oxidation state of uranium, e.g. the uranyl cation UO<sub>2</sub>(2+), is thermodynamically favored. For example, in U ore deposits in which uraninite - consisting of reduced U(IV) as UO<sub>2</sub>(2+x) - is the parent material, exposure to oxidizing conditions results in alteration to U(VI) minerals, with the U(VI)-phosphates frequently defining the boundaries of the ore body. U(VI)-phosphates are of interest because of their relatively low solubilities compared to other U(VI) solid phases. Since microorganisms are undoubtedly present in such ore deposits, they likely play a role in the formation of U(VI)-phosphate solid phases. To assist the U.S. Department of Energy (DOE) with long-term stewardship issues associated with bioremediation of uranium, the overall goal of this project is to work with model biological systems to define the mechanisms by which microorganisms facilitate the formation of U(VI)-phosphate solid phases. This information can then be used by DOE to design remediation systems that stimulate biological activity to favor the formation of U(VI)-phosphate phases.

In this project, we are investigating the role of some individual bacterial strains (*Bacillus sphaericus* and *Shewanella putrefaciens*) as well as microbial consortia isolated from the NABIR Field Research Center at Oak Ridge National Laboratory on the alteration of U(VI) solid phases. These strains were selected to reflect a variety of subsurface conditions including aerobic, microaerophilic, and episodically anaerobic. These bacteria or similar species are found throughout subsurface environments. They are believed to influence actinide geochemistry through various mechanisms. These mechanisms are not independent of one another, and together they illustrate the dynamic life cycle that defines the biogeochemical cycle of U(VI).

## B22A-0145 1330h POSTER

**The Biological Nature of Geochemical Proxies: algal symbionts affect coral skeletal chemistry**Kate Owens<sup>1</sup> (keowens@midway.uchicago.edu)Anne L Cohen<sup>1</sup> (508 289 2958; acohen@whoi.edu)Nobu Shimizu<sup>1</sup> (508 289 2963; nshimizu@whoi.edu)<sup>1</sup>Department of Geology and Geophysics, Woods Hole Oceanographic Institution, Quissett Campus, Woods Hole, MA 02543, United States

The strontium-calcium ratio (Sr/Ca) of reef coral skeleton is an important ocean temperature proxy that has been used to address some particularly controversial climate change issues. However, the paleothermometer has sometimes proven unreliable and there are indications that the temperature-dependence of Sr/Ca in coral aragonite is linked to the photosynthetic activity of algal symbionts (zooxanthellae) in coral tissue. We examined the effect of algal symbiosis on skeletal chemistry using *Astrangia danae*, a small colonial temperate scleractinian that occurs naturally with and without zooxanthellae. Live symbiotic (deep brown) and asymbiotic (white) colonies of similar size were collected in Woods Hole where water temperatures fluctuate seasonally between -2°C and 23°C. We used a microbeam technique (Secondary Ion Mass Spectrometry) and a 30 micron diameter sampling beam to construct high-resolution Sr/Ca profiles, 2500 microns long, down the growth axes of the outer calical (thecal) walls. Profiles generated from co-occurring symbiotic and asymbiotic colonies are remarkably different despite their exposure to identical water temperatures. Symbiotic coral Sr/Ca displays four large-amplitude annual cycles with high values in the winter, low values in the summer and a temperature dependence similar to that of tropical reef corals. By comparison, Sr/Ca profiles constructed from asymbiotic coral skeleton display little variability over the same time period. Asymbiotic Sr/Ca is relatively insensitive to the enormous temperature changes experienced over the year; the temperature dependence is similar to that of nighttime skeletal deposits in tropical reef corals and non-biological aragonite precipitates. We propose that the large variations in skeletal Sr/Ca observed in all symbiont-hosting coral species are not related to SST variability per se but are driven primarily by large seasonal variations in skeletal calcification rate associated with symbiont photosynthesis. Our model provides a framework for understanding the role of biology in determining coral skeletal chemistry and an explanation for anomalous Sr/Ca-based paleotemperature derivations.

## B22B MC: Hall D Tuesday 1330h

**Long Term Survival of Geologically Sequestered Microorganisms: Biogeological Conditions and Implications (joint with P)****Presiding:** R Holt, The University of Mississippi; T Kieft, New Mexico Tech; D Powers, Consulting Geologist

## B22B-0146 1330h POSTER

**Limit for the Survivability from Potassium Decay of Bacterial Spores in Halite Fluid Inclusions**Gerhard Kminek<sup>1</sup> (1-858-534-2995; gkminek@ucsd.edu)Jeffrey L Bada<sup>1</sup> (1-858-534-2995; jbad@ucsd.edu)<sup>1</sup>Scripps Inst. Oceanography, UCSD, 9500 Gilman Drive, La Jolla, CA 92093-0208, United States

Vreeland *et al.*<sup>1</sup> recently claimed to have isolated and cultured a viable spore forming halotolerant bacterium from a 250 million year old brine inclusion present in a salt crystal from the Salado formation. An earlier report suggested that viable bacterial spores could be revived from samples obtained from insects entombed in 25-40 million year old Dominican amber<sup>2</sup>. On the bases of these reports, Parkes<sup>3</sup> raised the question of whether bacterial spores under some conditions might be effectively immortal.

Sporulation, induced by an adverse change in the environmental conditions, is able to stabilize the DNA primarily against hydrolytic depurination for extended periods of time<sup>4</sup>. However, the organism is still exposed to ionizing radiation from the environment. Dormant spores have a reduced sensitivity to ionizing radiation per se, but unlike active organisms are unable to repair DNA damage encountered during long-term

exposure to ionizing radiation. The accumulated damage may overwhelm any repair mechanism that starts in the early stage of spore germination<sup>5</sup>.

The main radionuclide in a halite fluid inclusion is <sup>40</sup>K, which accounts for 0.0117% of natural potassium. <sup>40</sup>K decays via beta decay to <sup>40</sup>Ca and via electron capture to <sup>40</sup>Ar, releasing a primary gamma-ray. About 83.3 % of the beta's emitted are in the energy range of 0.3-1.3 MeV.

We assume 7 g/l for an average concentration of natural potassium in a halite fluid inclusion, which means that the amount of <sup>40</sup>K in a 10 μl fluid inclusion is 8.19 ng. We have chosen a 10 μl because this volume is typical of that used to obtain chemical data and in the attempts to extract bacteria. Less than a percent of the gamma decay energy is absorbed in a fluid inclusion of 10 μl. Thus, we will not take the gamma decay energy into account for the further discussion. Almost all the beta energy is absorbed in the fluid inclusion. The total decay energy absorbed in a time period of 250 million years is about 87 kGy.

The most DNA damage-tolerant organism known today is *Deinococcus radiodurans*. The viability of *D. radiodurans* falls to undetectable levels<sup>6</sup> at about 18 kGy. The survival curve of dry *Bacillus megaterium* spores shows a 4-log reduction at about 8-10 kGy<sup>7,8</sup>. These numbers can be compared to the 87 kGy in the case for a Permian fluid inclusion. The ability to tolerate radiation induced damage lies in the efficient repair mechanism employed by *D. radiodurans*, which is not operative in a dormant spore. It would thus be highly unusual for a bacterial spore to survive intact for 100's of millions of years unless these bacteria are extremely radiation tolerant.

Based on these considerations and without active DNA repair mechanisms, viability of a dormant bacterial spore and the survival of viable genetic material over extended periods of geologic time is probably limited by exposure to natural background radiation.

1. Vreeland, R. H., Rosenzweig, W. D., Powers, D. W. Nature 407, 897-900 (2000) 2. Cano, P. J., Borucki, M. K. Science 268, 1060-1064 (1995) 3. Parkes, R. J. Nature, 407, 844-845 (2000) 4. Lindahl, T. Nature 362, 709-715 (1993) 5. Setlow, P. J. Bacteriol., 174 (9), 2737-2741 (1992) 6. Battista, J. R. Annu. Rev. Microbiol. 51, 203-224 (1997) 7. Powers, E. L., Kaleta, B. F. Science 132, 959-960 (1960) 8. Ewing, D., Powers, E. L. Science 194, 1049-1051 (1976)

## B22B-0147 1330h POSTER

**Recycling Sequestered Bacteria Through Natural Hydrogeologic Processes**Dennis W. Powers<sup>1</sup> (915-877-3929; rmholt@olemiss.edu)Robert M. Holt<sup>2</sup> (662-915-6687; rmholt@olemiss.edu)Erik L. Powers<sup>3</sup> (erikpowers99@hotmail.com)<sup>1</sup>Consulting Geologist, 140 Hemley Road, Anthony, TX 79821, United States<sup>2</sup>The University of Mississippi, Department of Geology and Geological Engineering, 118 Carrier Hall, University, MS 38677, United States<sup>3</sup>Colorado State University, Department of Microbiology, Ft. Collins, CO 80523, United States

A report of viable Permian-age bacteria (sp. 2-9-3) preserved in salt has been contested on grounds that the revived bacteria are not genetically different enough from modern bacteria to be that old. Mutation rates are assumed to apply over geologic periods to this bacterium, but this hypothesis is not testable without reference to geologically old organisms. Recycling organisms by natural processes could complicate such schemes.

The Permian Salado Formation at the bacterium-sampling site has been protected well from natural sources of contamination. Elsewhere the unit has been subject to dissolution for at least 10-20 Ma potentially liberating trapped bacteria. Where the formation approaches the surface, it becomes much thinner and develops a solution residue on top of undissolved salt. At Nash Draw, a shallow valley is developing by solution and collapse, brine collects on and in the residue and flows as groundwater toward the Pecos River. Brine springs discharge into the Pecos River, which joins the Rio Grande, and ultimately reaches the Caribbean. Unless the sample from the Salado contained the only viable bacterium in the formation, 2-9-3 has likely been reintroduced into the biosphere frequently, if not nearly continuously, over millions of years. For such situations, unraveling the genetic relationships and mutation history will be more complicated than currently assumed.

## B22B-0148 1330h POSTER

**Deep-Subterranean Microbial Habitats in the Hishikari Epithermal Gold Mine: Active Thermophilic Microbial Communities and Endolithic Ancient Microbial Relicts.**Hisako Hirayama<sup>1</sup> (+81-468-67-5556; hirayamah@jamstec.go.jp)Ken Takai<sup>1</sup> (+81-468-67-3894; kent@jamstec.go.jp)Fumio Inagaki<sup>1</sup> (+81-468-67-9687; inagaki@jamstec.go.jp)Koki Horikoshi<sup>1</sup> (+81-468-67-5540; horikok@jamstec.go.jp)

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Deep subterranean microbial community structures in an epithermal gold-silver deposit, Hishikari gold mine, southern part of Kyusyu Japan, were evaluated through the combined use of enrichment culture methods and culture-independent molecular surveys. The geologic setting of the Hishikari deposit is composed of three lithologies; basement oceanic sediments of the Cretaceous Shimanto Supergroup, Quaternary andesites, and auriferous quartz vein. We studied the drilled core rock of these, and the geothermal hot waters from the basement aquifers collected by means of the dewatering system located at the deepest level in the mining sites. Culture-independent molecular phylogenetic analyses of PCR-amplified ribosomal DNA (rDNA) recovered from drilled cores suggested that the deep-sea oceanic microbial communities were present as ancient indigenous relicts confined in the Shimanto basement. On the other hand, genetic signals of active thermophilic microbial communities, mainly consisting of thermophilic hydrogen-oxidizer within Aquificales, thermophilic methanotroph within  $\alpha$ -Proteobacteria and yet-uncultivated bacterium OPB37 within  $\beta$ -Proteobacteria, were detected with these of oceanic relicts from the subterranean geothermal hot aquifers (temp. 70-100°C). Successful cultivation and FISH analyses strongly supported that these thermophilic lithotrophic microorganisms could be exactly active and they grew using geochemically produced hydrogen and methane gases as nutrients. Based on these results, the deep-subsurface biosphere occurring in the Hishikari epithermal gold mine was delineated as endolithic ancient microbial relicts and modern habitats raising active lithotrophic thermophiles associated with the geological and geochemical features of the epithermal gold deposit.

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**Hydrocarbon Gases in Hydrogeologically Isolated Fractures in Au Mines of the Witwatersrand Basin, South Africa: Potential Substrates for Deep Subsurface Microorganisms**Julie A.M. Ward<sup>1</sup> (1-416-978-0662;ward@geology.utoronto.ca); Greg F. Slater<sup>1</sup> (1-416-978-0825;slater@quartz.geology.utoronto.ca); Georges Lacrampe-Couloume<sup>1</sup> (1-416-978-0825;gcl@zircon.geology.utoronto.ca); James Hall<sup>2</sup> (1-609-258-1622; hall@Princeton.EDU); Duane Moser<sup>2</sup> (1-509-372-2098; duane.moser@pnl.gov); Li Hung Lin<sup>2</sup> (1-609-258-1622;lhlin@Princeton.EDU); Johanna Lippmann<sup>3</sup> (1-845-365-8514; Lippmann@ldeo.columbia.edu);Mark Davidson<sup>4</sup> (1-27-082 364 2637;davidson@gecko.biol.wits.ac.za); T.C. Onstott<sup>2</sup> (1-609-258-1622; tullis@Princeton.EDU); BarbaraSherwood Lollar<sup>1</sup> (1-416-978-0825;

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Deep subsurface microbial communities are the subject of a multi-disciplinary study in the Witwatersrand Basin, of South Africa. Hydrocarbon and H<sub>2</sub> gases found in the mines were investigated to determine their origin and role as potential substrates for long-term survival of microorganisms. Large quantities of gas (up to 30L/min/borehole) are released when sealed fracture systems are opened by exploration drilling. Two