Special Session

Fe-Oxidizing Microbial Observatory (FeMO)

Fall AGU Meeting 2007

Program for 18 and 19 December 2007

- **Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese I**

  B22D : Tuesday Morning 10:20 in MW 2006

- **Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese II**

  B23G : Tuesday Afternoon 13:40 in MW 2006

- **Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese III**

  B24D : Tuesday Afternoon 16:00 in MW 2006

- **Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese IV: Posters**

  B33A : Wednesday Afternoon 13:40 in MS Exh Hall B

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Session Summary

Iron and manganese are the most abundant redox active metals in Earth's crust, and thus are widely available for microbial energy generation. While Fe- and Mn-cycling microbes have long been recognized, recent work has provided great insight into their phylogenetic and physiological diversity, effects on environmental processes, roles in microbial ecology, and enzymatic pathways. In this session we aim to bring together researchers in geo- and biosciences to showcase the wide variety of lab-, field-, and computational-based approaches and to encourage dialogue between disciplines. Topics may include: What microbes oxidize and reduce Fe and Mn? What environments are they found in? What genes and proteins are involved? What is the geochemical/mineralogical state of Fe and Mn in the environment, and how do microbes affect this? What are tracers and biomarkers of Fe/Mn microbial activity and can we relate these to features in the rock record?

Conveners:

Chan, C and Orcutt, B.

About FeMO

The importance of metals to life has long been appreciated. Iron (Fe) is the fourth most abundant element and the second most abundant redox-active element in the Earth's crust, rendering it the most important environmental metal. Although a surge of research on microbial iron transformations has highlighted the importance of these processes, the majority of these studies have focused on the reductive Fe cycle and low pH environments, which arguably does not represent the global ubiquity of iron-rich habitats. The major purpose of the **Fe-Oxidizing Microbial Observatory (FeMO)** project is to elucidate the full physiological, phylogenetic and biochemical diversity of the long-neglected group of the environmentally relevant and ecologically important neutrophilic iron oxidizing bacteria. Our team selected the **Loihi Seamount** because it affords the maximal cross-section of diverse habitats over a minimal spatial scale, from low-flow, low-temperature environments to high-flow, high-temperature environments, as well as basaltic rock habitats.

Reduced (oxygen-poor) forms of iron (and possibly manganese) deep in the Earth and contained in volcanic rocks provide a rich source of energy for the growth of a very interesting group of rust-forming (iron-oxidizing) bacteria. Loihi Seamount is a hydrothermally active seamount in the mid-Pacific ocean where waters containing high concentrations of reduced iron exit the Earth's crust and where massive amounts of volcanic rock come in contact with sea water. Here abundant iron-oxidizing bacteria, in the darkness of the deep sea, carpet the rock surfaces, forming rust coatings on massive areas of the seamount's surface. The bacteria form microbial mats and crusts on exposed rock surfaces and greatly accelerate the alteration of these rocks and affect water chemistry. In turn, the rust (iron oxide) impacts other microorganisms and affects elemental cycling on the planet. Processes such as these are probably widespread in deep dark environments where water circulates below the surface of land and the oceanic crust. The primary focus of our FeMO project and this cruise is to understand who the important iron-oxidizing bacteria in the environment are, how fast they form iron oxide deposits, how they do it biochemically, and how this process affects ocean chemistry and ecosystem function.

Program

❖ Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese I

Tuesday Morning 10:20 in MW 2006

Time	Session	Title
10:20	**B22D**	**Presiding: Chan, C; Orcutt, B**
10:20	B22D-01 INVITED	The Diverse Microbiology of Anaerobic Fe(II) Oxidation **Coates, J D**; Weber, K A; Scherer, M; Achenbach, L A
1035	B22D-02	Anaerobic Nitrate-Dependent Metal Bio-Oxidation **Weber, K**; Knox, T; Achenbach, L A; Coates, J D
1050	B22D-03 INVITED	Constraining the role of anoxygenic phototrophic Fe(II)-oxidizing bacteria in deposition of BIFs **Kappler, A**; Posth, N R; Hegler, F; Wartha, E; Huelin, S
11:05	B22D-04	Physiology and Mechanism of Phototrophic Fe(II) Oxidation by Rhodopseudomonas palustris TIE-1 **Jiao, Y**; Newman, D
11:20	B22D-05	Biochemistry and Ecology of Novel Cytochromes Catalyzing Fe(II) Oxidation by an Acidophilic Microbial Community **Singer, S W**; Jeans, C J; Thelen, M P; VerBerkmoes, N C; Hettich, R C; Chan, C S; Banfield, J F
11:35	B22D-06	Expression of Genes Involved in Iron and Sulfur Respiration in a Novel Thermophilic Crenarchaeon Isolated from Acid-Sulfate-Chloride Geothermal Systems **Kozubal, M**; Macur, R; Inskeep, W P
11:50	B22D-07 INVITED	Biogeochemistry of Iron Oxidation in a Circumneutral Freshwater Habitat **Duckworth, O**; Homstrom, S; Pena, J; Zacharias, E; Sposito, G
12:05	B22D-08	Energy Filtering Transmission Electron Tomography (EFTET) of Bacteria-Mineral Associations within the Deep sea Hydrothermal Vent Shrimp Rimicaris exoculata **Anderson, L M**; Halary, S; Lechaire, J; Frébourg, G; Boudier, T; Zbinden, M; Laval, J; Marco, S; Gaill, F

❖ Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese II

Tuesday Afternoon 13:40 in MW 2006

Time	Session	Title
13:40	**B23G**	**Presiding: Chan, C; Orcutt, B**
13:40	B23G-01 INVITED	Neutrophilic Iron-Oxidizing Microbes In The Marine Environment **Edwards, K J**; Chan, C; Orcutt, B
13:55	B23G-02	Spatial and Temporal Variability in Microbial Communities from Pre- and Post-Eruption Microbial Mats Collected from Loihi Seamount, Hawaii: An Update **Moyer, C L;** Davis, R E; Curtis, A C; Rassa, A C
14:10	B23G-03	Iron Cycling at Loihi Seamount **Emerson, D**
14:25	B23G-04	Biogeochemical Cycling of Iron Isotopes at Loihi Seamount **Rouxel, O J**; Edwards, K J; Moyer, C L; Wheat, G
14:40	B23G-05	Widespread Iron Oxidizing Bacterial Communities in a late Paleoproterozoic Marine Environment **Planavsky, N**; Rouxel, O; Bekker, A
14:55	B23G-06	Microbially Mediated Glass Alteration in the Geological Record: Textural clues for Microbial Functions **Staudigel, H**; Furnes, H; McLoughlin, N; Banerjee; N
15:10	B23G-07	The Distribution and Stabilisation of Dissolved Fe in Deep-sea Hydrothermal Plumes **Bennett, S A**; Achterberg, E P; Connelly, D P; Statham, P J; Fones, G R; German, C R
15:25	B23G-08	Particulate Organic Carbon and Iron Speciation within Deep-Sea Hydrothermal Plumes **Toner, B M**; Fakra, S C; Manganini, S J; Moffett, J W; German, C R; Edwards, K J

❖ Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese III

Tuesday Afternoon 16:00 in MW 2006

Time	Session	Title
16:00	**B24D**	**Presiding: Chan, C; Orcutt, B**
16:00	B24D-01	Manganese Dependent Anaerobic Oxidation of Methane **Beal, E**; House, C
16:15	B24D-02	Carbon and Manganese Cycling in the Columbia River's Estuarine Turbidity Maxima in the South Channel **Bräuer, S L**; Kranzler, K; Tebo, B M
16:30	B24D-03	Diversity and As-adsorption properties of Mn(II)-oxidizing bacteria within tropical wetlands of the Mekong Delta **Ying, S C**; Kocar, B D; Fendorf, S; Francis, C A
16:45	B24D-04	The Integrated Field-Scale Subsurface Research Challenge Site (IFC) at Rifle, Colorado: Preliminary Results on Microbiological, Geochemical and Hydrologic Processes Controlling Iron Reduction and Uranium Mobility **Long, P E**; Banfield, J; Bush, R; Campbell, K; Chandler, D P; Davis, J A; Dayvault, R; Druhan, J; Elifantz, H; Englert, A; Hettich, R L; Holmes, D; Hubbard, S; Icenhower, J; Jaffe, P R; Kerkhof, L J; Kukkadapu, R K; Lesher, E; Lipton, M; Lovley, D; Morris, S; Morrison, S; Mouser, P; Newcomer, D; N'Guessan, L; Peacock, A; Qafoku, N; Resch, C T; Spane, F; Spaulding, B; Steefel, C; Verberkmoes, N; Wilkins, M; Williams, K H; Yabusaki, S B
17:00	B24D-05	Sulfidogenesis Controls on Ferrihydrite Transformation and Repartitioning of Sorbed Arsenic **Kocar, B D**; Fendorf, S
17:15	B24D-06	Assessing the Geochemical Reactivity of Fe-DOM Complexes in Lacustrine Sediments Using Nitroaromatic Probe Compounds **Hakala, J**; Fimmen, R L; Chin, Y; Agrawal, S G; Ward, C P
17:30	B24D-07	Electrochemical Characterization Shewanella oneidensis Mr-1 MtrABC **Nuester, J**; Ross, D E; Hartshorne, R S; Brantley, S L; Butt, J N; Richardson, D; Tien, M
17:45	B24D-08	Secondary Iron Mineral Formation by Shewanellae Using Different Carbon Sources **Salas, E C**; Kukkadapu, R K; Fredrickson, J K; Nealson, K H

❖ Geomicrobiology and Environmental Biogeochemistry of Iron and Manganese IV: Posters

Wednesday Afternoon 13:40 in MS Exh Hall B

Time	Session	Title
13:40	**B33A**	**Presiding: Chan, C; Orcutt, B**
13:40	B33A-0840	Magnetite Formation in Magnetospirillum Gryphiswaldense Studied With FORC Diagrams **Carvallo, C**; Hickey, S; Menguy, N; Faivre, D
13:40	B33A-0841	Measurement of Microbially Induced Transformation of Magnetic Iron Minerals in Soils Allows Localization of Hydrocarbon Contamination **Kappler, A**; Porsch, K; Rijal, M; Appel, E
13:40	B33A-0842	Solid-state Au/Hg Microelectrode for the Investigation of Fe and Mn Cycling in a Freshwater Wetland: implications for methane production **Ma, S**; Luther III, G W; Keller, J; Madison, A S; Metzger, E; Megonigal, J P; Emerson, D
13:40	B33A-0843	Seasonal Changes in the Biogeochemistry of Bacteriogenic Iron Oxides in a Groundwater-Supplied Wetland **Gault, A G**; Ibrahim, A; Langley, S; Renaud, R; Clark, I D; Fortin, D; Ferris, G
13:40	B33A-0844	Iron oxidation and biomineralization by Mariprofundus ferrooxydans, a deep-sea microaerophilic lithoautotroph **Chan, C S**; Emerson, D; Fakra, S; Edwards, K J
13:40	B33A-0845	In Situ Chemical Profiling of an Extremely low Temperature Hydrothermal System at Loihi Seamount, Hawaii **Glazer, B T**; Briggs, R A
13:40	B33A-0846	Zeta-Proteobacteria dominate the formation of microbial mats in low-temperature hydrothermal vents at Loihi Seamount **Rassa, A C**; McAllister, S M; Safran, S A; Moyer, C L
13:40	B33A-0847	Bacterial Diversity and Spatial Variability Found in a Mn-Fe Oxide Encrusted Microbial Mat From the 5000 Meter-Deep Hydrothermal Vent 'Ula Nui, Hawaii **Davis, R E**; Moyer, C L; Curtis, A C; Staudigel, H; Tebo, B M
13:40	B33A-0848	Dinitrogen Gas Production From Epi- and Endolithic Basalt Communities at Loihi Seamount: A New Challenge for the Marine Nitrogen Budget **Horn, G**; Kroeger, K D; Kartal, B; Jetten, M S; Edwards, K J
13:40	B33A-0849	Deciphering Life in Young Ocean Crust: Development and Testing of Methods for Subsurface Microbial Observatories and In-Situ Incubations **Orcutt, B N**; Edwards, K; Wheat, G; Jannasch, H
13:40	B33A-0850	Optimization of DNA Extraction from Deep-sea Basalt **Wang, H**; Edwards, K J
13:40	B33A-0851	The impact of hydrothermally-sourced Fe on ocean biogeochemistry: new investigations from the East Pacific Rise, 9-10°N **German, C R**; Manganini, S J; Rouxel, O J; Edwards, K J
13:40	B33A-0852	Aging of the Glass to Altered-Glass Boundary in Seamount Basalts **Fisk, M**; Tepley III, F; Keller, R
13:40	B33A-0853	Mineralization of Fe/Mn-precipitates From Hot Springs in the Sanbe Volcanic Area, Japan **Sakamoto, A**; Ishibashi, J; Kimura, H
13:40	B33A-0854	Diversity of Thermophilic Microorganisms within Hawaiian Fumaroles **Ackerman, C A**; Anderson, S; Anderson, C

13:40	B33A-0855	Evolution of the Microbial Community Structure and Iron Reduction Rate in a Column Biostimulation Experiment During the Transition From Iron to Sulfate Reduction **Jaffe, P R**; ElBishlawi, H; Hettich, R L; Kerkhof, L; Komlos, J; Kukkadapu, R P; Lipton, M S; Long, P E; McGuinness, L; Moon, H; Peacock, A D; VerBerkmoes, N C; Williams, K H
13:40	B33A-0856	Kinetics of Microbially Mediated Iron Reduction: The Role of Biomass, Ferrous Inhibition, and Electron Shuttles **MacDonald, L H**; Moon, H; Jaffe, P R
13:40	B33A-0857	Comparative Effects of Electron Transfer Mediators on the Bioreduction of Fe(III) Oxide **O'Loughlin, E J**
13:40	B33A-0858	Immobilization of Arsenic by Vivianite Biomineralization **Kim, H**; Park, B; Lee, I; Roh, Y
13:40	B33A-0859	Localisation and Functional Analysis of a Manganese Oxidase Protein From the Marine Alpha-Proteobacterium Aurantimonas manganoxydans (sp. SI85-9A1) **Anderson, C R**; Davis, R; Tebo, B
13:40	B33A-0860	Characterization of Microbial Communities in Coal Mine Drainage Treatment Systems With Elevated Manganese **Tan, H**; Zhang, G; Burgos, W
13:40	B33A-0861	Biodiversity of Rock Varnish at Yungay, Atacama Desert, Chile **Kuhlman, K**; Venkat, P; La Duc, M; Kuhlman, G; McKay, C
13:40	B33A-0862	Nutrient Dynamics and Carbon Microbial Mineralization Processes in the Pichavaram Intertidal Mangrove Sediments from India **Mathukumalli, B**
13:40	B33A-0863	Arsenite-Dependent Anoxygenic Photosynthesis by Bacteria from Mono Lake, California **Hollibaugh, J T**; Budinoff, C R
13:40	B33A-0864	Aerobic and Anaerobic Oxidation of Organic Acids in Yellowstone Hot Spring Ecosystems **Windman, T O**; Zolotova, N; Shock, E
13:40	B33A-0865	Clay-Bacteria Systems and Biofilm Production **Steiner, J**; Alimova, A; Katz, A; Steiner, N; Rudolph, E; Gottlieb, P
13:40	B33A-0866	Heavy Metal Contaminated Soils in Riverside Park, Milwaukee, WI: Character, Bioavailability, and Distribution **Dansand, J J**; Knudsen, A C

Abstracts

listing in alphabetical order

B33A-0854

Diversity of Thermophilic Microorganisms within Hawaiian Fumaroles

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Fumaroles provide heat and moisture characteristic of an environment suitable for thermophilic microorganisms. On the Island of Hawaii, fumaroles are scattered across the southeastern portion of the island as a result of the volcanic activity from Kilauea Crater and Pu'u' O'o vent. We used metagenomics to detect 16S rDNA from archaeal and bacterial thermophilic microorganisms indicating their presence in Hawaiian fumaroles. The fumaroles sampled exist along elevation and precipitation gradients; varying from sea level to 4,012ft and annual rainfall from less than 20in to greater than 80in. To determine the effects of environmental gradients (including temperature, pH, elevation, and precipitation) on microbial diversity within and among fumaroles, we obtained 22 samples from 7 fumaroles over a three-day period in February of 2007. Temperature variations within individual fumaroles vary from 2.3^{o}C to 35^{o}C and the pH variances that range from 0.4 to 2.0. Temperatures of the different fumaroles range from 29.9^{o}C to greater than 105^{o}C, with pH values that vary from 2.55 to 6.93. Further data on the microbial diversity within fumaroles and among fumaroles will be determined once the sequencing of the microbial 16S rDNA regions is completed. We are currently assembling and sequencing clone libraries of bacterial and archaeal 16S rDNA fragments from fumaroles.

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B33A-0859

Localisation and Functional Analysis of a Manganese Oxidase Protein From the Marine Alpha-Proteobacterium Aurantimonas manganoxydans (sp. SI85-9A1)

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The marine α-proteobacterium SI85-9A1 was isolated in 1985 from the oxic-anoxic transition zone in Sannich Inlet, Vancouver Island, Canada. This organism was originally isolated due to its ability to oxidise manganese from Mn(II) to Mn(IV) oxides. It later proved to have a functional Type I RuBisCO suggesting it can potentially grow autotrophically with Mn(II) acting as the electron donor. Until now the mechanism of Mn(II) oxidation has remained enigmatic although clues from the genome and preliminary experiments suggested SI85-9A1 possessed a manganese oxidase similar to the Mox oxidase produced by Pedomicrobium sp. ACM 3067. Whole cell fractionation techniques were used to localise this protein to the outer membrane where it is loosely bound and can be washed off. Manganese oxidation activity in non-pure crude extracts was analysed using an LBB colorimetric assay and monitoring Mn(III)-pyrophosphate complex formation at 258 nm in a Mn(III) capture assay. Both these assays not only confirmed function but suggested that this protein is extremely active with a manganese oxidation rate approaching 8.9 µM min-1 mg-1. Partial FPLC purification confirmed that this protein was approximately 50 KD in size and may be part of a larger holoenzyme.

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B22D-08

Energy Filtering Transmission Electron Tomography (EFTET) of Bacteria-Mineral Associations within the Deep sea Hydrothermal Vent Shrimp Rimicaris exoculata.

Anderson, L M; Halary, S; Lechaire, J; Frébourg, G; Zbinden, M; Gaill, F
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The chemical and temperature conditions around deep sea hydrothermal vents are both dynamic and extreme, yet the shrimp Rimicaris exoculata flourishes around these environments on the Mid--Atlantic Ridge (MAR). Epibiotic bacteria and minerals found within the branchial chamber (BC) of the shrimp are of great interest in the search for a chemical model for the Rainbow MAR hydrothermal vent site. Here we examine the close, three-- dimensional (3D) relationship between bacteria (on the inner surface of the BC wall) and the minerals that surround them. The morphology and chemistry of the minerals were analysed by Energy filtering Transmission Electron Microscopy (EFTEM, on a LEO--912 microscope) and X-ray Nano-analysis (EDXN, on a JEOL--2010 FEG microscope) respectively, and the 3D organization was determined by Transmission Electron Tomography (TET) and EFTET. Consecutive thin and semi--thin sections of 50--80nm (for EFTEM and EDXN) and 200--250nm (for TEM and EFTET) were cut through the BC cuticle and mounted on standard microscope grids. Sections were observed initially for morphology, to find broad relationships between bacteria and minerals. EFTET series acquisition was performed under cryo-conditions (-175°C) using a LEO-912 microscope. At each position of interest four tilt series were taken at two degree increments between -55° and +55° at various energy--losses: 1) zero--loss (ref); 2) 720 eV, 3) 690 eV and 4) 670 eV, to reconstruct the 3D location of iron. Tilted series were obtained using the ESIvision program (Soft--Imaging Software, Münster, Germany) with additional in--house scripts for automated acquisition. The 3D EFTET reconstruction volume was produced from the four tilted series using recently developed EFTET--J software (http://www.snv.jussieu.fr/~wboudier/softs.html). In many cases the observed minerals exhibit a sharp boundary against the bacteria, often with a substantial void between bacterial membrane/cell wall and mineral boundary. Mineral layering and zoning are also present. Our findings highlight the potential importance of iron as an energy source for Rimicaris exoculata epibionts at Rainbow, from their close association. The results from this study are contributing to the formulation of a chemical model for the Rainbow hydrothermal vent site (MAR).

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B24D-01

Manganese Dependent Anaerobic Oxidation of Methane

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Understanding the anaerobic oxidation is not only important for understanding hydrocarbon degradation but it also important for understanding the global carbon cycle. The anaerobic oxidation of methane (AOM) is a large sink for methane consuming 5-20% of today's methane flux (Valentine and Reeburgh, 2000), yet the requirements for this process are not well understood. It has been suggested that no other electron acceptors other than sulfate can be used in the AOM (Nauhaus, 2005). However, our new data suggests that manganese, in the form of birnessite, can be used as an electron acceptor instead of sulfate (Beal et al., in prep). Methane seep sediment from the Eel River Basin, CA was incubated with methane, 13C-labeled methane, and carbon dioxide. Because the net result of the AOM is the production of carbon dioxide from methane, the rate of the AOM in each of the incubations can be determined by measuring the incorporation of 13C in the carbon dioxide. Using this method, it was found that cultures incubated with nitrate showed inhibition of the AOM, while cultures incubated with iron gave inconclusive results. The only positive results that were found for alternate electron acceptors are the incubations that were given manganese and no sulfate, which showed methane oxidation. Further, when more manganese was injected into these incubations, the rate of AOM increased. Preliminary analysis of the microbial population using terminal restriction fragment length polymorphism (TRFLP) targeting the mcr gene showed an unidentified organism in these cultures. Future work with TRFLP, as well as clone libraries, will help to identify the organisms responsible for this process. Nauhaus, K., 2005, Environmental regulation of the anaerobic oxidation of methane: a comparison of ANME-I and ANME-II communities: Environmental microbiology, v. 7, p. 98. Valentine, D.L., and Reeburgh, W.S., 2000, New perspectives on anaerobic methane oxidation: Environmental Microbiology, v. 2, p. 477-484.

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B23G-07

The Distribution and Stabilisation of Dissolved Fe in Deep-sea Hydrothermal Plumes

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Iron (Fe) is an essential micronutrient for oceanic phytoplankton, yet the debate over its sources and sinks persists. Dissolved Fe(II) in hydrothermal vent fluids is enriched ca. 106-fold over open ocean values, but as vent-fluids enter the base of the water column, abundant polymetallic particulate phases are formed: predominantly Fe-rich sulfides and Fe oxyhydroxides. More recently in hydrothermal plumes, deviation from first order Fe oxidation kinetics has suggested the occurrence of organically stabilised dissolved Fe. Such stabilisation could have implications on the hydrothermal Fe flux to the deep-ocean. To study Fe stabilisation in non-buoyant hydrothermal plumes, we have investigated the plume system at 5°S, Southern Mid-Atlantic Ridge, for dissolved Fe concentrations and Fe-complexing ligands. Six CTD stations were occupied for this study that intercepted non-buoyant plumes. Along-axis flow dominated the dispersion of plume material within the ridge segment and, 2.5 km down-plume from the nearest vent-site, high concentrations of dissolved Fe (20 nM) were still present. These high levels of "dissolved" Fe could be due to the presence of Fe colloids and/or organic Fe complexes. With the use of Competitive Ligand Exchange- Cathodic Stripping Voltammetry (CLE-CSV), we have detected stabilised dissolved Fe complexes within this "dissolved" Fe fraction, on the edges of a plume. We calculate that such a stabilised Fe fraction in hydrothermal plumes, 2.9 to 5.6 times greater than deep-ocean dissolved Fe concentrations (0.7 nM), could be sufficient to provide 10-20% of the global deep-ocean dissolved Fe budget.

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B24D-02

Carbon and Manganese Cycling in the Columbia River's Estuarine Turbidity Maxima in the South Channel

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The Columbia River represents the largest input (60-90%) of fresh water to the California Current System, and provides a major source of dissolved manganese and nutrients to the coastal waters. Researchers have identified upper Estuarine Turbidity Maxima (ETM(s)) as hot spots for microbial activity, and it is here that extensive manganese cycling is thought to occur. Most probable number counts of microorganisms within the ETM have revealed that the cultivable numbers of manganese-oxidizing bacteria are not statistically significantly different than that of other heterotrophs when grown on defined media with simple carbon sources or low concentrations (0.05%) of casamino acids and were in the range of 103 - 104 cells per mL. Similar numbers of heterotrophs (9.3 X 103 cells/mL) were found using a nutrient-rich complex medium; however, the numbers of manganese-oxidizers were significantly lower (~13 cells/mL). Approximately 100 different manganese-oxidizing bacteria were isolated from different media and are being phylogenetically characterized. Measurements of dissolved, ascorbate-reducible and total Mn by inductively coupled plasma- optical emission spectroscopy revealed that concentrations of Mn are positively correlated with turbidity and thus are higher during an ETM event. In addition, dissolved, total, and ascorbate-reducible Mn were all negatively correlated with salinity, supporting the idea that the manganese originates in the river and is diluted by the seawater originating off the coast. Uptake of 14C-labeled bicarbonate in response to various electron donors (nitrite, ammonium, thiosulfate, or Mn(II)) was stimulated during an ETM event but not before or after, indicating that these electron donors may serve as potential energy sources for carbon fixation. Taken together, our results further demonstrate that ETMs are zones with high microbial activity and that the ETM microbial communities harbor the potential for carbon fixation even in the relatively nutrient-rich environment of the upper Columbia River estuary.

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B33A-0840

Magnetite Formation in Magnetospirillum Gryphiswaldense Studied With FORC Diagrams

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First-order reversal curve (FORC) diagrams, obtained from partial hysteresis curves, were measured on a set of cultured Magnetospirillum gryphiswaldense in order to study the formation of magnetite in magnetotactic bacteria. We used a cell-suspension assay for the growth-independent study of magnetite formation under highly controlled conditions. FORC diagrams showed a clear evolution from size distribution with a majority of superparamagnetic (SP) grains, to a distribution dominated by stable, single-domain grains, but still containing some SP particles. FORC diagrams also suggest that magnetostatic interactions increase during the formation and growth of the magnetite grains. TEM observations confirm this evolution. According to the saturation isothermal remanent magnetization (SIRM) cooling and warming curves, the Verwey transition can only be seen in the most mature samples, and well below 120 K. This suggests that the samples may have suffered from some partial oxidation

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B33A-0844

Iron oxidation and biomineralization by Mariprofundus ferrooxydans, a deep-sea microaerophilic lithoautotroph

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The ocean crust contains a large reservoir of reduced iron, available for microbial energy generation. Some of this ferrous iron is mobilized by fluids in hydrothermal fields at seamounts and mid-ocean ridges. A microaerophilic iron oxidizer, Mariprofundus ferrooxydans has been identified (by molecular methods and microscopy) at various sites, and appears to be a key iron-oxidizing bacterium (FeOB) in the deep sea. Originally isolated from microbial mats near vents at the Loihi Seamount in Hawaii, Mariprofundus is distinctive because it forms an extracellular iron-mineralized stalk-like structure. We aim to understand its metabolism and mineral formation using a multidisciplinary approach, including electron microscopy, x-ray spectroscopy, time-lapse light microscopic imaging of live cells, and genomic and biochemical analyses. Microscopy and spectroscopy work shows that as the cells grow, they excretes iron and organic-rich fibrils that make up the stalk, at a rate of ~2 microns/hr. Stalk growth appears to be parallel to the direction of Fe and oxygen gradients. The Mariprofundus genome contains several terminal oxidases/peroxidases, including two cbb3-type cytochrome oxidases with a high affinity for oxygen, consistent with the microaerophilic lifestyle of these organisms. However, we have not identified genes for metabolisms other than aerobic iron oxidation, nor have we found any genes similar to known or suspected iron oxidases, though the genome (2.87 Mb) is rich in cytochromes (32 of 2922 genes). Thus, we are performing experiments to extract and analyze proteins from both cultured and environmental samples in order to find ones that will oxidize iron. UV-Vis spectra of extracts suggest that c-type cytochromes are particularly abundant, so these are candidates for further investigation. In combination with the microscopy and spectroscopy studies, these are the first steps towards understanding the complete pathway of iron from uptake through mineral formation and growth.

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B22D-01; INVITED

The Diverse Microbiology of Anaerobic Fe(II) Oxidation

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Although anaerobic microbial oxidation of Fe(II) has been know for over a decade there is still a paucity of information available on this important metabolic process or the organisms involved. Recent studies have indicated that the metabolism is ubiquitous and a broad diversity of organisms are capable of oxidizing Fe(II) in the absence of oxygen. Our previous studies demonstrated the existence of geochemical conditions conducive to supporting the activity of nitrate-dependent Fe(II) oxidizing bacteria (NFoB) in sedimentary environments. As part of these studies we isolated and characterized several novel NFoBs. Three of these organisms, Diaphorobacter sp. strain TPSY, Ferrutens nitratireducens strain 2002 and Azospira suillum strain PS are currently undergoing whole genome shotgun sequencing in an effort to gain insight into the biochemistry and molecular biology of this geochemically important metabolism. These organisms represent diverse genera capable of anaerobically oxidizing Fe(II) using nitrate as the electron acceptor. Two of these organisms, strain 2002 and strain TPSY, are also capable of the anaerobic nitrate-dependent oxidation of U(IV) to U(VI). Diaphorobacter sp. strain TPSY was isolated from uranium and nitrate contaminated groundwater and is a member of the Comamonadaceae family in the beta subclass of the Proteobacteria, closely related to Diaphorobacter nitroreducens. It represents the first example of an anaerobic Fe(II)-oxidizer from the Comamonadaceae family and grows mixotrophically requiring an organic carbon source when growing with Fe(II) and nitrate as the electron donor and acceptor respectively. F. nitratireducens strain 2002 was isolated from aquatic sediment and is the type strain of a new genus, Ferrutens, in the beta class of the Proteobacteria. Its closest relative is Chromobacterium violaceum, a common soil bacterium. In contrast to C. violaceum, F. nitratireducens is non-fermentative and does not produce free cyanide (CN-) or the purple/violet pigments indicative of violacein production, a characteristic of Chromobacterium species. F. nitratireducens strain 2002 is currently the only described mesophilic autotrophic nitrate-dependent Fe(II)-oxidizing organism and biogenically produces green rust as a primary end-product of this metabolism. Azospira suillum strain PS is a member of the beta subclass of the Proteobacteria and was isolated based on its ability to grow anaerobically by dissimilatory perchlorate reduction from animal waste lagoons. It represents one of the dominant perchlorate-reducing genera in the natural environment. Although this organism does not grow by Fe(II) oxidation, phenotypic studies revealed its ability to couple Fe(II) oxidation to nitrate reduction and it was the first organism shown to be capable of the oxidation of the Fe(II) content of silicaceous Fe(II) minerals including almandine and staurolite. It was also the first organism shown to be capable of biogenic magnetite production through anaerobic Fe(II) oxidative processes. These varied organisms demonstrate both the phylogenetic and metabolic diversity of organisms involved in the anaerobic bio-oxidation of Fe(II) and raise many questions regarding the underlying mechanisms involved in this important metabolism in the natural environment.

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B33A-0866

Heavy Metal Contaminated Soils in Riverside Park, Milwaukee, WI: Character, Bioavailability, and Distribution

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Prior to being breached in 1990, the North Avenue Dam on the Milwaukee River had created a 2.5-mile impoundment for over 150 years. Upstream urban runoff and industrial pollution resulted in the deposition of heavy metal rich sediments in the slow moving waters of the impoundment. After the dam removal, the river returned to a more natural flowpath and as the river narrowed, newly exposed riverbed was annexed as part of Riverside Park, enabling ecological recovery efforts on the river and riparian zones. However, these newly exposed soils are enriched with heavy metal contaminants, most notably, Pb, Zn, Cd, Cu, and Ni, concentrated by the impoundment. The current study has analyzed the location and concentrations of these trace metals, as well as their mobility and availability. This study is being conducted in conjunction with the Urban Ecology Center, a nonprofit environmental organization located in Riverside Park that is dedicated to serving the local community and urban youth while restoring and protecting the natural areas along the Milwaukee River. Analyses have included determination of general soil parameters such as particle size, organic content, and point of zero charge analyses. Beyond bulk chemical analysis, we have conducted selective sequential extractions to estimate the chemical speciation of these elements, which showed that approximately 30 percent of contaminants are highly available. Additionally, the soils have been analyzed with an Electron Microprobe to

directly observe phase relationships of metals in the soils. Microprobe and other analyses have shown that heavy metals are associated with a variety of phases, including Mn and Fe oxy-hydroxides, and vary in concentration and phase relationships with depth and distance from the river. Finally, a field-portable x-ray fluorescence spectrometer (pXRF), coupled with GPS data, is being used to create a geochemical map of heavy metal distributions throughout the park.

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B33A-0847

Bacterial Diversity and Spatial Variability Found in a Mn-Fe Oxide Encrusted Microbial Mat From the 5000 Meter-Deep Hydrothermal Vent 'Ula Nui, Hawaii

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'Ula Nui Vent Field was discovered on the southern flank of Loihi Seamount during the 2006 FeMO Microbial Observatory expedition at over 5000 meters depth. The vent field exhibited abundant low temperature (<2°C above ambient) seeps which were covered with extensive mineral-encrusted microbial mat material. The microbial mats consisted of a 0.5-3 cm thick upper mat which was comprised of laminated layers of manganese oxides and iron oxides which overlayed a flocculent iron oxide mat that could attain depths of over 1 m deep. Bacterial communities from the top and bottom mats were analyzed using SSU rRNA terminal-restriction fragment polymorphisms (T-RFLP) coupled with traditional clone library analysis. T-RFLP chromatograms indicate dominance of the ζ- Proteobacteria in both the top and lower mat. Cluster analysis of the T- RFLP fingerprints show a strong correlation between the bottom mat and iron oxide-encrusted microbial mats found in the hydrothermally active Pele's Pit near the summit of Loihi Seamount. The top mat clusters with iron and manganese oxide encrusted microbial mats found at various sites on Loihi Seamount not associated with measurable active hydrothermal venting. Clone library analysis show that the top mat was dominated by phylotypes related to the δ- γ- and the recently described ζ- Proteobacteria, along with members of the Planctomycete Division. The dominance of ζ- Proteobacteria and Planctomycete phylotypes implies that neutrophilic iron oxidation and anaerobic ammonia oxidation are active metabolisms in the top mat bacterial community. Anaerobic ammonia oxidation coupled with nitrite reduction may also be an integral metabolism in this community since some Planctomycete phylotypes from the top mat cluster within the anammox clade.

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B22D-07; INVITED

Biogeochemistry of Iron Oxidation in a Circumneutral Freshwater Habitat

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Iron(II) oxidation in natural waters at circumneutral pH, often regarded as an abiotic process, may be biologically- mediated when it occurs in iron-rich redox gradients. West Berry Creek, a small circumneutral tributary flowing through a mixed coniferous forest in Big Basin State Park, California, contains localized iron (hydr)oxide precipitates at points along its course where anoxic groundwater meets oxygenated creek water. These mixing zones establish redox gradients and iron-rich microbial mats that may create microenvironments that promote active biogeochemical cycling of Fe. Water sampling revealed strong correlations between the concentrations of aqueous inorganic species, suggesting a rock-weathering source for most of these solutes. Liquid chromatography-mass spectrometry detected significant concentrations of organic exudates, including low molecular mass organic acids and siderophores, indicating active biogeochemical cycling of iron is occurring in the creek. X-ray diffraction and elemental analysis showed the iron precipitates to be

amorphous minerals, such as ferrihydrite. Microbial biofilm communities are associated with the iron (hydr)oxide deposits. Clone libraries developed from 16s rDNA sequences revealed the presence of microorganisms related to the neutrophilic iron- oxidizing bacteria Gallionella and Siderooxidans; in addition, micrographs suggest the widespread presence of sheath-forming bacteria (e.g., Leptothrix). Sequences from these libraries also indicated the presence of significant populations of organisms related to bacteria in the genera Pseudomonas, Sphingomonas, and Nitrospira. These geosymbiotic systems appear to be significant not only for the biogeochemical cycling of iron in the creek, but also for the cycling of organic species, inorganic nutrients, and trace metals.

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B23G-01; INVITED

Neutrophilic Iron-Oxidizing Microbes In The Marine Environment

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Neutrophilic iron-oxidizing bacteria (FeOB) have been recognized, described, enriched for and isolated from terrestrial aquatic and soil habitats for over one hundred years. Microbiologists and geoscientists alike have appreciated the important role FeOB play in processes such as corrosion and mineral deposition. However, recognition of their role and activities has lagged considerably behind in marine realms. Over approximately the past twenty years, however, there has been mounting interest and recognition of the presence and ubiquity of marine FeOB, particularly in the deep sea . Their role in rock and mineral weathering and alteration, mineral deposition, and biomass production in the deep sea has come into focus and is the subject of intense study. This paper will present an overview of marine FeOB including discussion of their habitats, diversity, and role in geochemical processes. In an effort to specifically target this elusive class of microbes, an observatory project for the study of FeOB and the marine microbial iron cycle has recently been launched--the Iron Microbial Observatory "FeMO", at the Loi'hi seamount, Hawaii. FeMO and FeOB isolated from Loi'hi are being used as model systems and testing grounds for studying mineral deposition and rock colonization in laboratory/field settings.

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B23G-03

Iron Cycling at Loihi Seamount

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Loi"hi is an active submarine volcano located close to the island of Hawai"i at a depth around 1100 m. The summit is host to a variety of focused and diffuse flow hydrothermal vent sites, which have in common a chemistry that is dominated by the presence of Fe(II) often in concentrations of 10's to 100's of μM at circumneutral pH. An outcome of these conditions is the deposition of substantial quantities of rust-colored microbial mats that are dominated by Fe-oxidizing bacteria (FeOB). These mat communities were first sampled in the late 1980s, and have subsequently been the subject of continued investigation, to the extent that Loih'i is now a model system for understanding deep-sea microbial communites that are primarily driven by iron-based lithotrophic metabolism. While the role of FeOB is becoming well-established, the short-term fate of the iron oxyhydroxides that are produced is less well understood, and specifically that role that Fe-reducing bacteria may have in re-cycling Fe(III) to Fe(II). A recent cruise to Loihi in 2006 found that one vent site, which has been slowly decreasing in activity, was essentially "dead", i.e. there was no sign of fluid flow or temperature anomaly. Yet, iron mats several cm thick persisted and chemical analysis suggested O2 penetrated the mat and that no Fe(II) was present, i.e. there no sign of active Fe-reduction. This raises the question of how active is the iron cycle in Loih'i iron mats? Experiments to measure Fe- reduction potential in Lohi"i mats do show activity and it is possible to enrich Fe-reducing bacteria. We have recently isolated a novel Geothermobacter sp. HR-1 from a Loih'i mat, that is an Fe-reducer and grows at mesophilic temperatures. This work will report on continued investigations into the iron cycle at Loih'i to further constrain the chemical conditions, further elucidate the potential for Fe-reduction in natural mat samples, and present the outcome of growing native isolates of Fe- oxidizers and reducers from Loih'i in co-culture.

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B33A-0852

Aging of the Glass to Altered-Glass Boundary in Seamount Basalts

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In oceanic basalts, fresh volcanic glass reacts with seawater, resulting in the loss of cations from the silica network and the formation of altered glass that retains insoluble cations and potassium. Microorganisms are associated with the glass to altered-glass (GAG) boundary, although their role is not known. It has been hypothesized that microorganisms promote the alteration of volcanic glass to take advantage of the reducing power of iron and manganese in the silicate glass, or alternatively, that microorganisms colonize the GAG boundary to use iron and manganese that are released during alteration. We examined the chemical composition of the GAG boundary of seamount basalts from the Gulf of Alaska that span the age range of <0.1 Ma to 49 Ma. All samples in the study were collected by manned or unmanned submersibles from outcrops on the sea floor, and were presumably exposed to bottom seawater from the time of eruption to the time of collection. The underlying assumptions of this study are that (1) the glass would become less reactive as it aged due to the closing off of fluid pathways as fractures became filled and as the surface of the basalt became coated with manganese crust; (2) the diminished fluid movement would result in the extinction of the original community of microorganisms, or in a change in the microbial community at the GAG boundary; and (3) the changing fluid flow and biological conditions in the rocks would result in changes in the composition of the altered glass at the GAG boundary. The younger basalts (<7 Ma) have the granular texture at the GAG boundary that has been associated with microorganisms. The older samples in this study (30 Ma and 49 Ma) have no, or only minor, granular texture. The altered glass of the older samples have higher Ti and lower Al, Mg, and Mn than the altered glass of the younger samples. Fe is enriched in the altered glass relative to the glass in samples of all ages. Phosphorus, an element that could be elevated by the presence of microorganisms, is only enriched in one area of altered glass in the 4 Ma sample. The aging of the GAG boundary does result in changes in composition of the altered glass, however, the influence, if any, of microorganisms on this change is not known.

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B33A-0843

Seasonal Changes in the Biogeochemistry of Bacteriogenic Iron Oxides in a Groundwater- Supplied Wetland

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Bacteriogenic iron oxides (BIOS), composed of a mixture of poorly ordered hydrous ferric oxides and intact and partially decomposed bacterial cells, are thought to play an important role in regulating aqueous trace element concentrations in groundwater-supplied wetland environments. We have initiated a study examining the seasonal variation in the biogeochemistry of BIOS and its role in contaminant sequestration in a wetland site at Chalk River, Ontario, Canada, an area that hosts an exceptional abundance of BIOS. Surficial BIOS and subsurface cores were collected in spring (April) and summer (July) for microbial and bulk geochemical characterization. In addition, dialysis `peepers` samplers were deployed to examine the changes in selected dissolved elements with depth. SEM inspection of representative BIOS samples found a predominance of sheaths similar to Leptothrix spp., Fe(II)-oxidizing microbes known to thrive in the circumnetural pH, high dissolved Fe(II) (up to 5 mg L-1) and low dissolved oxygen (<5 mg L-1) environment found at the groundwater springs of our study site. Helical stalks reminiscent of iron-oxidizing Gallionella spp. were also occasionally observed. The BIOS mineralogy was dominated by 2-line ferrihydrite, although interestingly samples collected further downstream from the groundwater discharge zone appear more closely related to 6-line ferrihydrite. Iron reduction was noted at depth in both the sedimentary and porewater profiles, with the dissolved Fe(II) peak in the latter occurring at shallower depths in summer compared to the spring. Strontium, a contaminant of concern at this site, showed little seasonal variation, but its concentration was broadly correlated with that of dissolved iron and manganese. In addition, porewater strontium concentrations were 2-3 fold higher at a nearby control site where BIOS was absent, indicating the likely importance of BIOS in attenuating migration of this contaminant.

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B33A-0851

The impact of hydrothermally-sourced Fe on ocean biogeochemistry: new investigations from the East Pacific Rise, 9-10°N.

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We have initiated a new study as part of NSF's Ridge 2000 program to investigate the fate of dissolved Fe(II) released from high-temperature hydrothermal venting on the East Pacific Rise and its potential impact on local deep-ocean biogeochemical budgets. Our principal focus is on the biogeochemical cycling of Fe and the role of microbial activity in that cycle: Fe is released in abundance from high-temperature vents, it plays a major role in modifying the gross flux of other elements from hydrothermal vents to the oceans and hydrothermal sources have the potential to dominate the global deep ocean budget of dissolved Fe: an essential micronutrient. To achieve our goals we have collected samples from close to vent-sites at 9-10°N on the East Pacific Rise over a period extending from late 2004 to late 2007 that brackets an episode of recent volcanism (Winter 2005-2006). Thus, we are in a position to investigate both the "steady state" input from a deep-ocean vent-system and also how that system has been perturbed following volcanic activity out to a period of 1-2 years post-eruption. Sampling has been conducted using time-series sediment traps deployed on long-term moorings that have sampled particles settling from buoyant and non-buoyant plumes at 9°50`N and at a contrasting (sulfide- free) vent-site at 9°30`N. Our project is nested within a larger study (LADDER) that spans across disciplines from segment-scale ocean circulation in the 9-10°N area to larval transport between adjacent vent sites. Our specific on-going work includes: i) analyses for major and trace element flux variations; ii) investigation for Fe isotope fractionation within our hydrothermal plume samples; and iii) a study of the role of Fe- oxidising bacteria in catalysing Fe oxidation and/or Fe isotope fractionation within these hydrothermal plumes. For example, the mineralogical structure and character of Fe oxyhydroxide minerals may, themselves, result from the microbial catalysis of Fe oxidation.

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B33A-0845

In Situ Chemical Profiling of an Extremely low Temperature Hydrothermal System at Loihi Seamount, Hawaii

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Loihi Seamount is a submarine, active volcano located on the southeast flanks of the Big Island of Hawaii. It is considered to be the youngest volcano in the chain, sharing the hot spot magma chamber with Mauna Loa and Kilauea. Sites of both vigorous and diffuse hydrothermal venting can be found surrounding the pit crater summit (1000m) and on the flanks of the seamount, down to its base (5000m). Vent fluids at Loihi are chemically distinct from other well-studied marine hydrothermal systems and have been shown to be enriched in carbon dioxide, iron(II), and manganese(II), and deplete in sulfur species. The Loihi summit is located within a zone of low oxygen, further enabling elevated iron(II) concentrations and support for a dominant community of iron-oxidizing bacteria. We deployed a sensor wand consisting of up to four voltammetric working electrodes and the ROV Jason temperature probe, and/or a submersible micromanipulator with voltammetric electrodes to provide real time in situ redox characterizations of hydrothermal fluids and geochemical gradients associated with iron-oxidizing microbial mats and flocs. In addition to surveying known areas of warm temperature (10-60 degrees C) venting at the Loihi summit, we performed widespread profiling of a previously undescribed site at 5000m, that exhibits temperature anomalies of just 0.2 degrees C. Extensive iron-oxidizing microbial mats were shown to occur up to 2m in thickness over several hundred square meters. Bottom water oxygen concentrations were near-saturation, and we observed steep gradients at the mat-interface, with little oxygen penetration and iron(II) concentrations of up to 150 micromolar. Our in situ electrochemical analyses provided an efficient and valuable means for directed discrete sampling of hydrothermal fluids and microbial flocs, as well as previously unattainable high spatial resolution geochemical profiles through the mats.

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B24D-06

Assessing the Geochemical Reactivity of Fe-DOM Complexes in Lacustrine Sediments Using Nitroaromatic Probe Compounds

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The complexation environment of Fe in anoxic sediment porefluids is important for understanding its role in biogeochemical reactions in these systems. These include the microbial dissolution of iron-bearing sediments and the degradation of hydrophobic pesticides. We studied the effect of naturally occurring Fe-DOM complexes on the reduction of two nitroaromatic compounds (NACs) as a surrogate for probing electron-transfer kinetics, and monitored the geochemical parameters affecting reactivity. We found that Fe(II) was necessary for rapid NAC reduction (< 24 hr), and observed faster reduction with increased pH. NAC reduction in `pH-adjusted' porefluids (acidified to pH 2.5 after porefluid extraction and raised to the native pH (between 6.5 to 7.8) prior to reaction) was similar to that observed in model systems containing Fe(II) and aquatic fulvic acids. Conversely, NAC reduction in fresh, unaltered porefluids was slower than that observed in `pH-adjusted' porefluids, indicating that the natural metal redox speciation differs between fresh and `pH-adjusted' samples. Electrochemical scans demonstrate a vertical gradient in concentrations of Fe(III), Fe(II), and Mn(II) from 0 to 30 cm depth. Furthermore, the electrochemical data indicate that ferric iron in these systems is predominantly complexed with organic ligands, with a half-cell potential of -0.45 V. The magnitude of this ferric-organic peak decreases when porefluids are pH- adjusted for short durations (acidified to pH 2.5 and re-raised to circumneutral levels) and disappears completely when the acidified porefluid is allowed to equilibrate overnight. These data show that pH-adjustment of porefluids presumably alters both their complexation chemistry and reactivity towards NACs, and shows how small changes in Fe complexation chemistry potentially affects electron transfer reactions in anoxic environments.

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B33A-0863

Arsenite-Dependent Anoxygenic Photosynthesis by Bacteria from Mono Lake, California

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Chemolithoautotrophic growth of bacteria capable of using inorganic arsenic compounds is well documented. Energy conservation and carbon fixation have been demonstrated in both arsenite oxidizers and arsenate reducers from many bacterial lineages under both aerobic and anaerobic conditions. However, phototrophic growth using reduced arsenic as an electron donor has not been described. Here we report the light-dependent oxidation of arsenite to arsenate, coupled with autotrophic growth, by an enrichment culture of bacteria from Mono Lake, California. The culture is dominated by a phototrophic member of the Ectothiorhodospiraceae that is >99% similar to Ectothiorhodospira sp. (strain Borgoria Red). It also contains an uncultured member of the Bacteriodetes and another member of the Ectothiorhodospiraceae that is 96% similar to Alkalilimnicola erhlichii. The bacteria grow more slowly and clump when grown on arsenite versus sulfide or thiosulfate. Scanning electron micrographs reveal that they are closely associated and enmeshed in an organic matrix.

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B33A-0848

Dinitrogen Gas Production From Epi- and Endolithic Basalt Communities at Loihi Seamount: A New Challenge for the Marine Nitrogen Budget

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Current knowledge of the nitrogen cycle in the oceans indicates a net loss of biologically available nitrogen from the oceans. One marine habitat that is conspicuously absent from current nitrogen budget estimations is seafloor-exposed lavas, such as occur at young ridge flanks around mid-ocean ridge systems and seamounts. Basalt from two locations on Loihi Seamount, an actively venting volcano southeast of the island of Hawai'i, were incubated shipboard with 15N labelled nitrate and ammonium additions to determine the rate of nitrogen loss mediated by epi- and endolithic microbial communities. In all incubations a net production of dinitrogen gas occurred with concurrent consumption of oxidized nitrogen species (nitrate plus nitrite) and ammonium. The availability of reduced iron and manganese from the basalts could serve as potential electron donors for microbial metabolisms, including denitrification and anaerobic ammonia oxidizing (anammox) bacteria. The presence of 16S rRNA gene sequences that align with the anammox clade of Planctomycetes and ongoing anammox enrichment cultures inoculated with Loihi basalt suggest a role for anammox bacteria as well. This initial study indicates an active and significant role for basalt associated microbial communities in the nitrogen budget of the oceans.

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B33A-0855

Evolution of the Microbial Community Structure and Iron Reduction Rate in a Column Biostimulation Experiment During the Transition From Iron to Sulfate Reduction

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During the biostimulation of iron reducers for the purpose of concurrent biological reduction of U(VI), it has been postulated that iron reduction proceeds while bioavailable iron is present, after which the system switches to sulfate reduction if sulfate is present. Field experiments from the Rifle Integrated Field Challenge (IFC) site in Colorado showing that the onset of sulfate reduction has been associated with decreased removal of U(VI) from groundwater support this hypothesis. However, column experiments using sediments from the Rifle site and synthetic groundwater with comparable (7 mM) sulfate levels as in the field, showed that the onset of sulfate reduction occurred within a month with no negative effect on U(VI) reduction. Separate column experiments using low (9 uM) sulfate concentrations showed that iron reduction can be maintained for over 200 days with no indication of iron limitations. To address the discrepancy between field and column experiments, an experiment is being conducted to determine the activity of iron reducers before and after the onset of sulfate reduction. Since Fe(II) buildup is difficult to quantify in the presence of sulfate reduction, the sediments were augmented with Fe- 57 goethite. Minute changes in the Fe-57 goethite can be detected via Mössbauer spectroscopy. Ten columns (2.5 cm internal diameter and 15 cm in length), loaded with sediment from the Rifle site, have been set up and are being operated at 17 °C. Groundwater from the Rifle site, amended with 3mM acetate and 20 μM U(VI), is pumped through the columns at a rate of 0.035 ml/min. Column effluent concentrations are being monitored for acetate, Fe(II), U(VI), and sulfate. Columns are sacrificed at 10 day intervals and the sediment samples are analyzed for Fe(II), U(IV), and acid volatile sulfides using standard analytical procedures. Changes in Fe-57

goethite measured using Mössbauer spectroscopy during biostimulation of the native microorganisms at 10-day intervals are showing measurable decreases in the goethite over these time intervals. Initial characterization of the microbial community in the sediment via DGGE of 16S rDNA shows the presence of Geobacter, Rhodoferax, Flexibacter, Flavobacterium, and Desulfuromonaceae. Very similar microbial populations were observed in column effluents using TRFLP analysis. Changes in individual bacterial cell numbers are being quantified via qPCR, while changes in bacterial activity are being quantified via proteomics.

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B22D-04

Physiology and Mechanism of Phototrophic Fe(II) Oxidation by Rhodopseudomonas palustris TIE-1

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Phototrophic Fe(II)-oxidizing bacteria use electrons from ferrous iron [Fe(II)] and energy from light to drive reductive CO2 fixation. This metabolism is thought to be ancient in origin, and plays an important role in environmental iron cycling. It has been implicated in the deposition of Banded Iron Formations, a class of ancient sedimentary iron deposits. Consistent with this hypothesis, we discovered that hydrogen gas, a thermodynamically favorable electron donor to Fe(II), in an Archean atmosphere would not have inhibited phototrophic Fe(II) oxidation. To understand this physiology and the connection to BIF formation at the molecular level, the mechanisms of phototrophic Fe(II) oxidation were examined in a model organism Rhodopseudomonas palustris TIE-1. Increased expression of a putative decaheme c-type cytochrome, encoded by pioA, was observed when cells were grown under Fe(II)-oxidizing conditions. Two genes located immediately downstream of pioA in the same operon, pioB and pioC, encode a putative outer membrane beta-barrel protein and a putative high potential iron-sulfur protein, respectively. Deletion studies demonstrated that all three genes are involved in phototrophic Fe(II) oxidation. This study provides our first insight into the molecular mechanisms of this metabolism, which will be further characterized by in vitro biochemical studies.

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B22D-03; INVITED

Constraining the role of anoxygenic phototrophic Fe(II)-oxidizing bacteria in deposition of BIFs

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Banded Iron Formations (BIFs) are Precambrian sedimentary deposits of alternating iron oxide and silica mineral layers. Their presence in the rock record ca.3.8-2.2 Ga makes them particularly intriguing formations for the debate over when oxygen became dominant on Earth. The mechanism(s) of BIF deposition is still unclear; suggestions including both abiotic and biotic processes. We are interested in constraining one of these proposed mechanisms; the direct biological oxidation of Fe(II) via anoxygenic Fe(II)-oxidizing autophototrophs. In order to find the limitations of photoferrotrophic BIF deposition, we take a holistic approach, investigating the oxidation of Fe(II) by modern Fe(II)-oxidizing phototrophs, the precipitation of Fe(III) (hydr)oxides, and the fate of the cell-mineral aggregates in the water column and at the basin floor. Specifically, physiology experiments with Fe(II)-oxidizing phototrophs under various conditions of light intensity, pH, Fe(II) concentration and temperature allow us to determine the environmental limits of such organisms. We carry out precipitation experiments to characterize the sedimentation rates, aggregate size and composition in order to resolve the effect of reactions in the water column. Finally, we simulate the diagenetic fate of these aggregates on the basin floor by placing them in gold capsules under T and P conditions relevant for the Transvaal Supergroup BIFs of South Africa. Recently, we have developed a tank simulating the Archean ocean in which the strains grow in continuous culture and collect the aggregates formed under various geochemical conditions. We aim to model the extent of and limitations to photoferrotrophs in BIF deposition. This information will help constrain whether biotic processes were dominant in the Archean ocean and will offer insight to the evolution of the early biogeosphere.

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B33A-0841

Measurement of Microbially Induced Transformation of Magnetic Iron Minerals in Soils Allows Localization of Hydrocarbon Contamination

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Soil contamination by crude oil and other hydrocarbons represents a severe environmental problem, but often the location and extent of contamination is not known. Hydrocarbons, or their degradation products, can stimulate iron-metabolizing microorganisms, leading to the formation or dissolution of (magnetic) iron minerals and an associated change of soil magnetic properties. Therefore, the screening of soil magnetic properties has the potential to serve as an efficient and inexpensive tool to localize such contaminations. In order to identify the influence of different biogeochemical factors on the microbially influenced changes of magnetic iron minerals after hydrocarbon contamination, oil spills were simulated in laboratory batch experiments. The parameters tested in these experiments included soils with different bedrocks, type and amount of added hydrocarbon, and microbiological parameters (sterile and autochthonous microorganisms). In order to follow the changes of the soil magnetic properties, the magnetic susceptibility of the samples was measured weekly. First results show that changes in the magnetic mineralogy are caused by microbial activity, as sterile samples showed no changes. In the microbially active set-ups, the magnetic susceptibility increased or decreased up to 10% in comparison to the initial magnetic susceptibility within a few weeks. In one iron-rich soil even a decrease of the magnetic susceptibility of ~40% was observed. Although the amount and type of hydrocarbons did not effect the changes in magnetic susceptibility, DGGE fingerprints revealed that they influenced microbial communities. These results show that the magnetic susceptibility changes in the presence of hydrocarbons and that this change is microbially induced. This suggests that the screening of soil magnetic properties can be applied to localize and assess hydrocarbon contamination. In order to understand the biogeochemical processes better, the change of the iron mineralogy will be followed by Moessbauer spectroscopy in future batch experiments. Furthermore, iron-metabolizing microorganisms are currently isolated and identified.

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B33A-0858

Immobilization of Arsenic by Vivianite Biomineralization

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Microbial metal reduction and biomineralization has the potential for immobilizing metals and radionuclides in subsurface environments. The objective of this study was to examine Fe reduction and biomineralization in the presence of arsenite or arsenate using metal-reducing bacteria, Shewanella sp. (Haejae-1), enriched from an intertidal flat sediment, South Korea. The bacteria was able to use glucose as an electron donor and Fe(III)-citrate as an electron acceptor in the presence of arsenite or arsenate using phosphate buffered saline medium. The reduction of Fe(III)-citrate in the presence of arsenite or arsenate resulted in the precipitation of white µm-sized crystalline minerals. XRD analysis of the white precipitate after 14-day incubation identified the mineral phase as vivianite. SEM with EDX analysis of the vivianite precipitated by the metal reducing bacteria confirmed the presence of Fe, As, O, and P. This study indicates that formation of sparingly soluble vivianite precipitates, mediated by the metal -reducing bacteria, may sequester iron, phosphate, and arsenic into more stable and less toxic forms. The formation of phosphate minerals has frequently been observed in sedimentary environments under high biological productivity, where organic matter serves as a source of phosphate to sediment pore water through bacterial degradation. Therefore, formation of sparingly soluble iron precipitates, mediated by the metal- reducing bacteria, may sequester iron, phosphate, and other metals into more stable and less toxic forms in subsurface environments.

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B24D-05

Sulfidogenesis Controls on Ferrihydrite Transformation and Repartitioning of Sorbed Arsenic

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Iron (hydr)oxides are ubiquitous sorbents of arsenic (As) that undergo reductive dissolution and transformation upon reaction with dissolved sulfide. Here, we examine diverging pathways of solid phase iron (Fe) transformation during sulfate reduction in the presence of varying As loadings. Columns initially containing As(V)- ferrihydrite coated sand, inoculated with the sulfate reducing bacteria Desulfovibrio vulgaris (Hildenborough), were eluted with artificial groundwater containing sulfate and lactate. Additionally, abiotic batch reaction experiments were conducted to examine Fe secondary products rapidly formed during sulfidization of As-loaded ferrihydrite. Rapid and consistent sulfate reduction coupled with lactate oxidation is observed within column solids possessing low As(V) surface coverage (10% of the adsorption maximum). Column experiments illustrated that at high As(V) surface coverage (50% of the adsorption maximum), sulfate reduction and lactate oxidation are initially slow but gradually increase over time, and all As(V) is reduced to As(III) by the end of experimentation. The dominant Fe solid-phase transformation products at low As coverage include amorphous FeS within the zone of sulfate reduction (near the inlet of the column) and magnetite downstream where Fe(II)aq concentrations exceed 1 mM. Arsenic(V) is reduced to As(III) and displaced from the zone of sulfidogenesis and Fe(III)s depletion. At higher As coverage, green rust carbonate, as opposed to magnetite, is a dominant Fe solid phase product. Independent of loading, As is strongly associated with magnetite and residual ferrihydrite, while being excluded from green rust and iron sulfide. Abiotic batch reactor experiments illustrate that As is readily released from ferrihydrite during sulfidization, and that low As loadings yield initial Fe secondary products of lepidocrocite and FeS, while high loadings inhibit rapid secondary Fe mineral formation. Our observations illustrate that sulfidogenesis occurring in proximity with Fe (hydr)oxides may govern pathways of Fe solid phase transformation and As partitioning; formation of As sulfide minerals, in particular, is inhibited by reactive Fe either through sulfide oxidation or complexation.

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B22D-06

Expression of Genes Involved in Iron and Sulfur Respiration in a Novel Thermophilic Crenarchaeon Isolated from Acid-Sulfate-Chloride Geothermal Systems

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Acidic geothermal springs within Yellowstone National Park (YNP) provide an excellent opportunity to study microbial populations and their relationship with geochemical processes such as redox cycling and biomineralization of iron. Fourteen acid-sulfate-chloride (ASC) and acid-sulfate (AS) geothermal springs located in (YNP) have been extensively characterized for aqueous chemistry, solid phase mineral deposition and microbial diversity and distribution. The oxidation of Fe(II) with oxygen as an electron acceptor is exergonic under these conditions, consequently, Fe(II) may be an important electron donor driving primary production in ASC and AS habitats, and products of biomineralization (e.g. Fe[III]-oxides of varying crystallinity and structure, as well as jarosite in some cases) are common in the outflow channels of these environments. Recently, we isolated a novel Metallosphaera-like microorganism (Metallosphaera strain MK1) from an ASC spring in Norris Geyser Basin, YNP. Clone libraries (16S rRNA gene) from multiple sites suggest that microorganisms closely related to strain MK1 (between 98-100 percent similarity) dominate many spring locations between 55-80 C. The in situ abiotic oxidation rate of Fe(II) has been shown to be very slow in these systems and Metallosphaera strain MK1 has been directly implicated in biotic Fe(II) oxidation. Metallosphaera strain MK1 has been submitted for full genome sequencing and is yielding gene sequences related to the terminal oxidases SOXABC and SOXM super-complex. In addition, sequences from a recently characterized terminal oxidase FOX complex involved in Fe(II) and pyrite oxidation from Sulfolobus metallicus have been found in Metallosphaera strain MK1. A protein

complex analogous to Metallosphaera sedula has been identified in strain MK1 and this complex has also been expressed in cells grown on pyrite and Fe(II). Other sequences identified in Metallosphaera strain MK1 that are involved in respiration are the TQO complex (thiosulfate:quinone oxidoreductase) related to the Acidianus ambivalens DOXAD complex and a sulfur reductase (SRE) complex related to one found in Sulfolobus solfataricus and Acidianus ambivalens. Here we report on the RNA expression of seven gene sequences from each of the above mentioned complexes for Metallosphaera strain MK1 grown aerobically on pyrite, sulfur, Fe(II)-ferrihydrite, and anaerobically with yeast extract and sulfur. In addition, expression studies are also compared to in situ samples collected from the geothermal Fe-mats.

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B33A-0861

Biodiversity of Rock Varnish at Yungay, Atacama Desert, Chile

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Rock varnish is a very slow-growing nanostratigraphic coating consisting of approximately 70% clay and 30% iron and manganese oxides of fine-grained clay minerals rich in manganese and iron oxides, which forms on the surfaces of rocks in most semi-arid to hyper-arid climates. Rock varnish has even been postulated to exist on Mars based on surface imagery from several landed missions, and is considered a potential biomarker. However, the mechanism of varnish nucleation and growth remains unknown. Whether or not microbes are involved in the nucleation and growth of rock varnish, the detection of microbes using cultivation or cultivation- independent techniques has demonstrated that varnish provides a microhabitat for microbes. We hypothesized that rock varnish in the Mars-like Yungay region of the Atacama Desert may provide such a microhabitat for microbial life where none has been found to date in the surface soil (< 1 cm). The presence of microbes was investigated using adenosine triphosphate (ATP) assay techniques and culture-independent biomolecular methods. High levels of both total and intracellular ATP were associated with the rock varnish while negligible ATP was found in the surrounding surface soil, suggesting that viable organisms were present. Total DNA was extracted from ground varnish and surrounding surface soil and subjected to trifurcate polymerase chain reactions (PCR). No DNA was recovered from the soil. Amplicons were used to generate ribosomal DNA (rDNA) clone libraries, which suggest the presence of numerous phylogenetically distinct microorganisms in eight Eubacterial clades, Alphaproteobacteria, Betaproteobacteria, Gammaproteobacteria, Cytophaga-Flavobacterium- Bacteroides (CFB), Chloroflexi (green non-sulfur bacteria (GNS)), Gemmatimonadetes, Actinobacteria and Cyanobacteria. The diversity of bacteria found and presence of cyanobacteria suggests that rock varnish provides a niche environment for a cryptoendolithic microbial community where life is unable to colonize the surface soil in a Mars-like environment.

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B24D-04

The Integrated Field-Scale Subsurface Research Challenge Site (IFC) at Rifle, Colorado: Preliminary Results on Microbiological, Geochemical and Hydrologic Processes Controlling Iron Reduction and Uranium Mobility

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The IFC at Rifle, Colorado was recently funded by the U.S. Department of Energy to address knowledge gaps in 1) geochemical and microbial controls on stimulated U(VI) bioreduction by iron-reducers, 2) U(VI) sorption under Fe-reducing conditions, 3) post-biostimulation U(VI) stability and removal, and 4) rates of natural bioreduction of U(VI). The over-arching goal of the project is to develop a mechanistic understanding of bioreductive and abiotic processes that control uranium mobility targeting new knowledge that can be translated into scientifically defensible flow and reactive transport process models. The Rifle IFC will conduct a focused set of field and lab experiments that use recently developed sciences of proteogenomics and stable isotope probing to track microbial metabolic status during acetate amendment. This information will be linked to changes in Fe redox status and sulfide minerals, with field-scale changes detected by non-invasive hydrogeophysics, including 3-D resistivity tomography. A key goal of the project is to combine abiotic sorption processes under reducing conditions with biotic processes controlling U(VI) reduction. The initial field-scale experiment for the Rifle IFC was conducted during the summer of 2007 with the objectives of collecting simultaneous metagenomic and proteomic samples during acetate amendment and to assess the impact of intentionally decreasing electron donor concentration on the metabolic processes of iron reducers. The 2007 experiment replicated previous field experiments, producing dominance of Geobacter sp. in groundwater within 10 days after the start of acetate amendment. The experiment also confirmed the importance of heterogeneities in controlling the flux of electron donor and the impact of naturally reduced zones on the duration of Fe reduction.

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B33A-0842

Solid-state Au/Hg Microelectrode for the Investigation of Fe and Mn Cycling in a Freshwater Wetland: implications for methane production

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The solid-state voltammetric gold-amalgam microelectrode was used to measure multiple redox species (O, S, Fe and Mn) in situ at (sub)millimeter vertical resolution to elucidate rhizosphere processes in Jug Bay wetlands. In vegetated soil, a classic diagenetic redox sequence without any dissolved sulfide was observed in summer. However, the rhizosphere can be quite variable which is due to the introduction of O2 to the anoxic sediments by plants. In non-vegetated soil, the vertical concentration-depth profiles were relatively constant. The presence of Fe(II), Mn(II) and soluble Fe(III) in deeper sediments indicates the oxidation of Fe(II) as well as the non-reductive dissolution of Fe(III) and the reductive dissolution of Fe(III) and Mn(III, IV) solids. Mn(III, IV) and Fe(III) redox chemistry is important in organic matter mineralization mediated by bacteria and in suppressing methane formation. In addition, Mn(III, IV) also can oxidize Fe(II) to supply Fe(III) for bacterial Fe(III) reduction. Studying Fe and Mn cycling via voltammetric methods can give insights to methane production and loss as there is no methane sensor for sediment work at present.

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B33A-0856

Kinetics of Microbially Mediated Iron Reduction: The Role of Biomass, Ferrous Inhibition, and Electron Shuttles

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Microbially mediated reduction of poorly crystalline iron oxides in the subsurface controls the fate and transport of many organic and inorganic contaminants and plays a role in biogeochemical nutrient cycling. Despite recent advances in understanding this important process, much remains undiscovered. In particular, the influence of biomass on iron reducing kinetics is not well understood for surface bound microbes undergoing direct electron transfer to ferric iron oxides. Nor do we understand how Fe(II) sorption to ferric iron influences this process under direct electron transfer versus transfer via redox shuttles. Accordingly, this study investigates the iron reducing kinetics and mineralogy of geobacter sulfurreducens reducing poorly crystalline iron coated sand using acetate as electron donor, with and without AQDS, an electron shuttle and humic analog. This research demonstrates that the rate of iron reduction depends logarithmically on initial biomass for surface bound geobacter, suggesting surface area limitations with increasing biomass. However, electron shuttles remove these limitations and shift the kinetics such that the rate depends linearly on biomass over the range of cell concentrations explored. This study also finds that ferrous iron inhibits the rate of reduction with or without AQDS, but has a weaker effect in the presence of AQDS, and adding an Fe(II) specific chelator removes the negative feedback mechanism altogether.

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B33A-0862

Nutrient Dynamics and Carbon Microbial Mineralization Processes in the Pichavaram Intertidal Mangrove Sediments from India

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Various nutrient parameters of the mangrove water were analyzed for one year (July 2003-June2004) to delineate the driving forces behind the biogeochemical nutrient dynamics in the Pichavaram mangrove waters of south east coast of India. With few exceptions, the over all trend was higher concentrations of nutrients in monsoon and post-monsoon periods. Regression analyses between salinity and various

nutrients explain that salinity had firm control over the nutrient distribution in the mangrove water. Stoichiometric ratios deviated greatly from the Redfield ratio, reflecting nutrient behavior that was non-conservative because of long residence time and high primary productivity associated with organic matter decomposition. Microbial nutrient mineralization rates were estimated at two different locations (i.e. impacted and un-impacted). Sulfate reduction is the major microbial mineralization pathway followed by metal (Fe-Mn) and aerobic decomposition. Total carbon oxidation (TCOX) were calculated and found that TCOX rates were higher in the pristine A. marina zone than in the two impacted stands (43 and 79 mol C m−2 y−1); rates of total carbon oxidation in the R. apiculata forest averaged 75 mol C m−2 y−1. The sulfate reduction was peaked at the surface to a depth of 1 m, leading to little carbon burial (~5% of total C input). The microbial activities are actively dampened by the anthropogenic activities and it alters the plant-soil relationship which ultimately influences the biogeochemical cycling of nutrients in the coastal environment.

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B23G-02

Spatial and Temporal Variability in Microbial Communities from Pre- and Post-Eruption Microbial Mats Collected from Loihi Seamount, Hawaii: An Update

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Loihi Seamount is an active submarine volcano that marks the southernmost extent of the Hawaiian hotspot. Loihi rises over 3000 meters from the seafloor and summits nearly 1000 meters below sea level. Hydrothermal activity was discovered at Loihi in 1987, yielding diffuse vent effluent (Tmax 37°C) with associated high CO2 and Fe(II) concentrations and luxuriant microbial mats located near the summit of the volcano. Loihi erupted most recently in 1996 forming a new 300 meter deep caldera (Pele's Pit) with hydrothermal venting up to 200°C. Pele's Pit has cooled and now contains multiple hydrothermal vents with hydrothermal fluids ranging from 8-58°C with concentrations of Fe(II) remaining between 50 and 750 µM. Community fingerprints from over 75 microbial mat samples have now been collected from Loihi Seamount from 1993 to 2006, with temperatures ranging from ambient (~4°C) up to nearly ~200°C. These samples were analyzed using Pearson product-moment coupled with UPGMA cluster analysis of terminal- restriction fragment length polymorphisms (T-RFLP) coupled with traditional clone library and sequence analysis to identify the primary populations within each community. These mat samples form two distinct community clusters (Loihi Cluster Group 1 and Group 2) representing a combined 90% of all mat samples collected. Loihi Cluster Group 1 is by far the largest group (n = 45) and contains the most mat samples collected over time. Group 1 is dominated by phylotypes closely related to the recently described zeta- Proteobacteria that includes the type strain Mariprofundus ferrooxydans, an obligately lithotrophic, Fe-oxidizing bacterium. Loihi Cluster Group 2 is comprised of only post-eruption communities (n = 18) that generally contain greater diversity (in terms of richness) than Group 1 communities. Group 2 communities are primarily dominated by a unique array of phylotypes belonging to the Nitrospira division and by the class epsilon- Proteobacteria, including many putative sulphur-oxidizing bacteria. Interestingly, we have recently witnessed a shift away from the Group 2 communities back to the Group 1 communities. Only 2 of 11 microbial mat samples collected in 2004 exhibited the Group 2 community structure. All 10 of the mat samples collected in 2006 from multiple locations exhibited the Group 1 community structure, indicating a reduced overall diversity within Pele's Pit and a return to the microbial mats being dominated by Fe-oxidizing bacteria. http://fire.biol.wwu.edu/cmoyer/research.html

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B24D-07

Electrochemical Characterization Shewanella oneidensis () Mr-1 MtrABC

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Dissimilatory iron-reducing bacteria have the ability to use a wide range of terminal electron acceptors including solid state iron (oxihydr)oxides. It is generally accepted that electrons are transferred by electron transfer proteins to a series of multiheme c-type cytochromes which enable the electron transport from the periplasm to the extracellular side of the outer cell membrane and across the bacteria-mineral interface to the terminal electron acceptor. In the last decade, the facultative anaerobe organism Shewanella oneidensis Mr-1 has been used as a model organism to identify, purify, and sequence single proteins involved in Fe(III) and Mn(IV) reduction, but these studies have provided little biochemical information on the actual electron transfer process within the bacterial cell. In order to extend the knowledge on electron transfer, Ross et al.(in press) have recently purified a complex from Shewanella oneidensis Mr-1 which includes the membrane proteins MtrA, MtrB, and MtrC and spans the space from the periplasm to the extracellular side of the outer membrane. In our study we applied the relatively new technique of protein film electrochemistry to the MtrABC complex to gain more biochemical information on electron transport in the membrane of Shewanella oneidensis Mr-1. A wealth of information on the reaction of redox-active sites in proteins like MtrABC can be acquired by voltammetric studies in which the protein sample is immobilized as a layer onto an electrode surface. By carrying out cyclic voltammetry over a wide range of scan rates, the data can be analyzed in terms of peak potentials versus scan rate. A simple reversible electron transfer process gives rise to a trumpet-shaped plot because the oxidation and the reduction peaks increasingly separate at high scan rate. In this contribution we show a detailed electrochemical picture of the MtrABC complex, which gives insight into the electron transfer from the periplasm to the extracellular side of the outer membrane of Shewanella oneidensis Mr-1. Such electrochemical analysis will help to understand how electrons are transferred to solid state electron acceptors such as ferrihydrite or goethite with different mineralogical and thermodynamic properties. Ross, D. E., Ruebush, S. S., Brantley, S. L., Hartshorne, R. S., Clarke, T. A., Richardson, D. J., and Tien, M., in press. Characterization of Protein/Protein Interactions Involved in Iron Reduction by Shewanella oneidensis MR-1. Applied and Environmental Microbiology.

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B33A-0857

Comparative Effects of Electron Transfer Mediators on the Bioreduction of Fe(III) Oxide

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The transfer of electrons from microbes to sparingly-soluble, extracellular electron acceptors such as Fe(III) oxides can occur via direct contact with the mineral surface, by dissolution of the mineral facilitated by exogenous or endogenous ligands and subsequent reduction of the dissolved Fe(III) ligand complex, and by facilitated electron transfer involving endogenous or exogenous electron transfer mediators (ETMs, also commonly referred to as electron shuttles) that are reduced by the microbes and then subsequently diffuse away from the cell and transfer electrons to the Fe(III) mineral surface, regenerating the oxidized form of the ETM. This study examines the effects of a series of compounds representing major classes of natural and synthetic organic ETMs (including low molecular-mass quinones, humic substances, phenazines, phenoxazines, phenothiazines, and indigo derivatives) on the bioreduction of lepidocrocite (γ-FeOOH) by the dissimilatory Fe(III)-reducing bacterium Shewanella putrefaciens CN32. S. putrefaciens CN32 was able to reduce lepidocrocite in the absence of exogenous ETMs; however, relative to the control, all of the synthetic ETMs examined in this study enhanced the bioreduction of lepidocrocite. The extent of the enhanced bioreduction increased with decreasing reduction potential of the given ETM redox couple.

However, the addition of Suwannee River fulvic acid, humic acid, or unfractionated NOM (10 mg organic C L-1) resulted in, at best, a minimal enhancement of lepidocrocite bioreduction relative to the control that did not contain any added exdogenous ETM. These results suggest that the relative contribution of humic substances to microbially mediated Fe(III) reduction may be minimal in low-carbon environments such as oligotrophic lakes and typical groundwaters.

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B33A-0849

Deciphering Life in Young Ocean Crust: Development and Testing of Methods for Subsurface Microbial Observatories and In-Situ Incubations

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Biogeochemical processes within the ocean crust, a hydrologically active global habitat, are hypothesized to balance key elemental budgets in the ocean (i.e. Si, Fe), yet the importance of microbial activity in mediating these processes remains unexplored. Reduced iron makes up roughly 10% of the basalt that comprises ocean crust, making iron cycling a likely source of energy for microbial activity in these systems. Available data indicates that young sub-seafloor ocean crust may be the most redox active: weathering features are established early (within the first 10-20 Ma) and change little thereafter. Laboratory studies, field examinations, and in-situ colonization and weathering experiments have shown that chemoautotrophic Fe- and S-oxidizing microorganisms are abundant and active in weathering rocks exposed at the seafloor at low temperatures. A current challenge is adaptation of available techniques for use in subsurface hard rock environments to evaluate the extent and activity of the deep biosphere. Because of the importance to study microbial communities under their relevant environmental conditions, significant resources must be dedicated to investigate this realm in situ. To address these challenges, sub-seafloor microbial observatories designed to yield high-quality, undisturbed samples that are amenable to genomic, taxonomic, spatial and geochemical profiling are in development and testing. Deployment of these observatories within drilled boreholes will allow the scientific community unprecedented opportunities to investigate microbial processes experimentally in sediments as well as subsurface hard rock environments. Initial experiments during ocean drilling expeditions at both the young and relatively warm Juan de Fuca flank as well as the young and cold ridge flank of North Pond along the Mid-Atlantic Ridge are crucial for evaluating whether the sub-seafloor ridge flank habitat supports a unique microbial community, how the microbial community compares in diversity and activity to those of deep sediments and seawater, and the global significance of microbes in biologically-mediated oxidative weathering of basalts.

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B23G-05

Widespread Iron Oxidizing Bacterial Communities in a late Paleoproterozoic Marine Environment

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Discovery of exceptionally well-preserved microfossils in the Gunflint Iron Formation provided the first evidence for diverse and abundant life in the Precambrian oceans. However, whether the Gunflint microbial communities were dominated by cyanobacteria or represent an archaic, lithotrophic ecosystem is a matter of longstanding debate. Here, we present Fe isotope and rare earth element (REE) data for microfossiliferous stromatolites that are consistent with the Gunflint biota being an iron-oxidizing bacterial community. The lack of or positive Ce anomalies in REE data indicate that the benthic community grew below a redoxcline and positive iron isotope compositions reveal that the microbial community only oxidized a fraction of the dissolved iron load. This depositional setting and Fe isotope evidence for incomplete iron oxidation are inconsistent with a cyanobacterial interpretation and provide strong support that the Gunflint biota was indeed a microaerophilic lithotrophic microbial ecosystem. An apparently restricted temporal distribution of the Gunflint biota coupled

with a widespread spatial distribution signals a significant ecosystem evolutionary event in the late Paleoproterozoic when the ocean was redox-stratified and Fe-rich anoxic waters impinged onto the shallow shelf.

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B33A-0846

Zeta-Proteobacteria dominate the formation of microbial mats in low-temperature hydrothermal vents at Loihi Seamount

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Loihi Seamount is Hawaii's youngest volcano and one of the earth's most active. Loihi is located 30 km SE of the big island of Hawaii and rises over 3000m above the sea floor and summits at 1100m below sea level. An eruption in 1996 of Loihi led to the formation of Pele's Pit, a 300 meter deep caldera. The current observations have revealed diffuse hydrothermal venting causing low to intermediate temperatures (10 to 65°C). The elevated temperatures, coupled with high concentrations of Fe(II) (ranging from 50 to 750 μM) support conditions allowing for extensive microbial mat formation. The focus of this study was to identify the colonizing populations of bacteria generated by the microbial mats at Loihi Seamount. Twenty-six microbial growth chambers were deployed and recovered after placement in the flow of hydrothermal vents for 3 to 8 days from within Loihi's caldera. Genomic DNA was extracted from samples and analyzed by Terminal-Restriction Fragment Length Polymorphism (T-RFLP) using eight restriction enzyme treatments to generate fingerprints from bacterial amplicons of small subunit rRNA genes (SSU rDNAs). Pearson product-moment coupled with UPGMA cluster analysis of these T-RFLP fingerprints showed that these communities bifurcated into two primary clusters. The first (Group 1) had an average vent effluent temperature of 44°C, and the second (Group 2) had an average vent effluent temperature of 64°C. Representative samples from within the two clusters (or groups) were chosen for further clone library and sequencing analysis. These libraries revealing a dominance of the recently discovered zeta- Proteobacteria in the lower temperature group (Group 1) indicating that they were the dominant colonizers of the microbial mats. These microaerophilic, obligately lithotrophic, Fe-oxidizing bacteria are most closely related to Mariprofundus ferrooxydans. The higher temperature group (Group 2) was dominated by epsilon- Proteobacteria primarily of the genus Sulfurimonas, which are sulfur- and thiosulfate-oxidizing bacteria. http://fire.biol.wwu.edu/cmoyer/research.html

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B23G-04

Biogeochemical Cycling of Iron Isotopes at Loihi Seamount

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It is now well recognized that seafloor hydrothermal systems support diverse and unique biological communities capable of using dissolved chemical species, such as Iron (Fe), as well as mineral substrates as sources of metabolic energy. Deep-sea hydrothermal systems such as the Loihi Seamount hydrothermal field are important examples of environments where both chemical and biological oxidation of Fe can occur simultaneously and provide an ideal system in which to test hypotheses on biotic vs. abiotic origin of iron-oxide formation. Here, we applied Fe isotope systematics of hydrothermal fluids and Fe-oxide precipitates to study biogeochemical cycling of iron and the formation of microbial mats at Loihi seamount. Warm hydrothermal fluids (<60°C) and iron oxide precipitates were recovered using the DSV Jason II during FeMO 2006 cruise. Fe-isotope composition of warm hydrothermal fluids yielded δ56Fe values near 0.1‰ and are indistinguishable from basalt values defined at 0.09‰. Suspended iron oxide particles in the fluids and seafloor iron oxide sediments (microbial mats) recovered in the vicinity of the vents yielded systematically positive δ56Fe values. The enrichment in heavy isotopes between 1.05 to 1.43‰ relative to Fe(II) in vent fluids is slightly higher than those obtained for abiotic Fe oxidation (around 0.9‰) and slightly lower than for bacterial Fe oxidation at circum neutral pH (around 1.5‰). Mass balance considerations also imply that the extent of Fe(II) oxidation is very limited in the vicinity of the vents (<20%) and that most Fe(II) is oxidized later in the water column. These results are consistent with the low oxygen content of seawater (i.e. summit of Loihi is located in the OMZ) and resultant slow kinetics of abiotic

Fe oxidation. In contrast, mats supported by very diffuse fluids recovered at the base of the Loihi Seamount (~ 5000m depth) have distinctly negative Fe-isotope values between -0.3 to -1.5‰. These negative values are best explained by near-complete oxidation of isotopically light Fe(II) source. Negative δ56Fe values in the source fluid are likely generated by subsurface precipitation of isotopically heavy Fe-oxides during partial Fe(II) oxidation. These results, together with the significant enrichment in Mn-oxides relative to the warm mats, are consistent with the higher oxygen level in deep seawater and suggest extensive microbial Fe(II) oxidation below seafloor. Fe-isotope compositions of microbial mats at Loihi Seamount display a remarkable range between -1.2 to 1.6‰ which enlarges considerably the range of δ56Fe values for other hydrothermal Fe-oxide deposits at mid-oceanic ridges (δ56Fe values between -0.8 to 0‰). This unique feature at Loihi indicates that Fe isotope compositions of hydrothermal Fe-oxide precipitates are particularly sensitive to oxygen levels in the local environment where they form, and are less sensitive to abiotic vs. biotic origins. The Loihi hydrothermal ecosystem provides an important modern analogue for testing hypotheses about the biogeochemical cycling of Fe-isotopes on early Earth.

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B33A-0853

Mineralization of Fe/Mn-precipitates From Hot Springs in the Sanbe Volcanic Area, Japan

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Mt. Sanbe is an active volcano located in western Japan. At southwest slope of Mt.Sanbe, hot springs are discharged with flow rate of 2000 liter per minute, which are accomapnied by iron and manganese precipitates. We studied chemical composition of hot spring waters by IC, ICP-AES and AA analysis, and of precipitates by XRF analysis. Water chemistry of the main hot spring (t= 30-39 °C) is Na-Cl type and contains significant iron (= 5.6-7.5 mg/l) and manganese (= 1.1-1.6 mg/l). Around the main spring discharge, both ferrihydrites (contains 67-76% of Fe) and manganese oxides (contains 71-79% of Mn) are precipitated. About 30 to 100m east from the main hot spring, black precipitates mainly consists of manganese oxides (contains 5-83% of Mn) are distributed in the area of 90 m × 45 m. Cold spring waters (t= 12-20 °C) in this area also contains Mn (0.1-0.6 mg/l), but their Fe concentration was under detection limit. Relative chemical comoposition of the cold spring waters are similar to that of the main hot spring, which implies dilution with groundwater. Total bacterial count measured by DAPI staining was 16,000-37,000 cells/ml in these waters. In order to estimate microbial activity, we conducted manganese oxidizing experiments. The oxidation rate was estiamted as 4-5 nM per hour for the experiment in which 5 g of fresh manganese wad was added to 200ml of the 0.20 μm filtered hot spring water. This rapid oxidation rate would suggest that microbial activity plays important role in manganese precipitation from the Sanbe hot/cold spring waters.

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B24D-08

Secondary Iron Mineral Formation by Shewanellae Using Different Carbon Sources

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It has been known for some time that microbes play an important role in the redox cycling of iron. A considerable amount of work has been done investigating the various factors that play a role in solid phase iron reduction such as the type of iron oxide available for reduction, medium composition, surface area and ferrous iron concentration. However, the rate at which a single cell reduces iron oxides and the impact of dissimilatory iron reduction kinetics on mineral speciation are not well understood. In order to determine the relationship between iron oxide reduction rates and secondary mineral formation, we have tested 4 different strains of Shewanella with different carbon sources. Strains MR-1, MR-4, CN32 and W3-18-1 were incubated with HFO (hydrous ferric oxide) as electron acceptor and either lactate, pyruvate, isoleucine or uridine as the organic carbon source. Mineral products were analyzed using X-ray diffraction, electron microscopy and Mossbauer spectroscopy. Initial results suggest that reduction rates are similar among strains when using the same carbon source.

However, qualitative assessment of mineral products suggests that while reduction rates for the tested strains may be similar, the secondary mineral products can be quite different.

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B22D-05

Biochemistry and Ecology of Novel Cytochromes Catalyzing Fe(II) Oxidation by an Acidophilic Microbial Community

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An acidophilic microbial community found in the Richmond Mine at Iron Mountain, CA forms abundant biofilms in extremely acidic ($pH<1$) and toxic metal conditions. In this ecosystem, biological Fe(II) oxidation is critical to the metabolic functioning of the community, and in turn this process generates acid mine drainage, causing an environmental catastrophe. Two conspicuous novel proteins isolated from these biofilms were identified as gene products of Leptospirillum group II and were characterized as cytochromes with unique properties. Sulfuric acid extraction of biofilm samples liberated one of these proteins, a 16 kDa cytochrome with an unusual alpha-band absorption at 579 (Cyt579). Genomic sequencing of multiple biofilms indicated that several variants of Cyt579 were present in Leptospirillum strains. Intact protein MS analysis identified the dominant variants in each biofilm and documented multiple N-terminal cleavage sites for Cyt579. By combining biochemical, geochemical and microbiological data, we established that the sequence variation and N-terminal processing of Cyt579 are selected by ecological conditions. In addition to the soluble Cyt579, the second cytochrome appears as a much larger protein complex of ~210 kDa predominant in the biofilm membrane fraction, and has an alpha-band absorption at 572 nm. The 60 kDa cytochrome subunit, Cyt572, resides in the outer membrane of LeptoII, and readily oxidizes Fe(II) at low pH (0.95 - 3.0). Several genes encoding Cyt572 were localized within a recombination hotspot between two strains of LeptoII, causing a large range of variation in the sequences. Genomic sequencing and MS proteomic studies established that the variants were also selected by ecological conditions. A general mechanistic model for Fe(II) oxidation has been developed from these studies. Initial Fe(II) oxidation by Cyt572 occurs at the outer membrane. Cyt572 then transfers electrons to Cyt579, perhaps representing an initial step in energy flow to the biofilm community. Amino acid variations and post-translational modifications of these unique cytochromes may represent fine-tuning of function in response to local environmental conditions.

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B23G-06

Microbially Mediated Glass Alteration in the Geological Record: Textural clues for Microbial Functions.

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Fe and Mn oxidizing microbes interact with their environment through the microbially mediated formation of Fe/Mn oxides and through the corrosion textures they may leave behind in the solids they colonize and from which they extract nutrients. Understanding the geobiology of Fe and Mn oxidation may focus on the study of the microbes themselves, the mineral products, its biocorrosion features and the relationships between these types of observations. We have reviewed our own data on glass bio-corrosion and in particular the wider literature on microbial mineral tunneling to develop a two stage biocorrosion model for volcanic glass that offers feedback for our understanding of the mechanisms and the dynamics of microbial dissolution. Traces of microbially mediated dissolution of volcanic glass are commonly observed in volcanic glass found in submarine volcanoes on the seafloor, and in uplifted submarine volcanoes of almost any geological age back to the origin of life. Two main bioalteration textures care observed, granular and tubular. Based on a comparison of these features in particular with tunneling by ectomycorrhizal fungi, we propose two distinct types of biocorrosion that affects glass: (1)

Granular alteration textures, made up of colonies of microbe-sized, near spherical mineral - filled cavities that form irregular clusters ranging to a tens of micron thick bands at the glas surfaces. These granular textures are interpreted as the result of microbial colonization. accompanied by dissolution of the glass in their contact surface, deposition of authigenic minerals and the formation of a biofilm, that eventually seals the glass from easy access by seawater for hydration, or from microbes accessing Fe (II) in the glass. (2) The most spectacular bioalteration feature, repesented by the formation of tubes cannot be easily formed by the former mechanism because near spherical, individual microbes are likely not to produce the directionality that is required to produce the near linear or sometimes coiled tubes. Instead, we envision the activity of hyphae-like organelles or filaments, that may radiate out from a host body located in direct contact with circulating water, possibly penetrating a biofilm and entering/drilling into the fresh glass. Such microdrilling is well described in soils, where hyphae can slowly drill into silicates, in a process that takes about 1000 years to become visible as tunnels.

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B33A-0865

Clay-Bacteria Systems and Biofilm Production

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Soil clots and the aerosol transport of bacteria and spores are promoted by the formation of biofilms (bacteria cells in an extracellular polymeric matrix). Biofilms protect microorganisms by promoting adhesion to both organic and inorganic surfaces. Time series experiments on bacteria-clay suspensions demonstrate that biofilm growth is catalyzed by the presence of hectorite in minimal growth media for the studied species: Gram negatives (Pseudomonas syringae and Escherichia coli,) and Gram positives (Staphylococcus aureus and Bacillus subtilis). Soil organisms (P. syringae, B. subtilis) and organisms found in the human population (E. coli, S. aureus) are both used to demonstrate the general applicability of clay involvement. Fluorescent images of the biofilms are acquired by staining with propidium iodide, a component of the BacLightTM Live/Dead bacterial viability staining kit (Molecular Probes, Eugene, OR). The evolving polysaccharide-rich biofilm reacts with the clay interlayer site causing a complex substitution of the two-water hectorite interlayer with polysaccharide. The result is often a three-peak composite of the (001) x-ray diffraction maxima resulting from polysaccharide-expanded clays and an organic-driven contraction of a subset of the clays in the reaction medium. X-ray diffractograms reveal that the expanded set creates a broad maximum with clay subsets at 1.84 nm and 1.41 nm interlayer spacings as approximated by a least squares double Lorentzian fit, and a smaller shoulder at larger 2q, deriving from a contraction of the interlayer spacing. Washing with chlorox removes organic material from the contracted clay and creates a 1-water hectorite single peak in place of the double peak. The clay response can be used as an indirect indicator of biofilm in an environmental system.

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B33A-0860

Characterization of Microbial Communities in Coal Mine Drainage Treatment Systems With Elevated Manganese

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Sediment samples were collected from two coal mine drainage treatment sites in western Pennsylvania. Both of the sites use constructed limestone beds to passively treat acidic coal mine drainage containing elevated manganese (Mn). Site #1 has influent manganese of 150 mg/L and effluent manganese between 40-100 mg/L. Site #2 has influent manganese of 20 mg/L and effluent manganese of less than 0.5 mg/L. Large quantities of black crusts were deposited throughout the beds at both sites. X-ray diffraction showed these crusts constituted of buserite, which is a layered structure manganese oxide mineral. Both culture-dependent and nucleic acid- based techniques were used to

characterize the bacterial and fungal communities in these beds. 16S rRNA gene analysis showed that bacterial communities were very diverse and included Cyanobacter, Proteobacteria, Bacteroidete, Planctomyceta, Acidobacter, Actinobacter and Gemmatimonade taxa. The archaeal diversity was lower and most sequences were related to uncultivated species. Two Mn-oxidizing fungi strains were isolated from one of the sites. One of the fungi is capable of oxidizing Mn(II) at both low and netural pH (3-7) while the other fungi can only oxidze Mn(II) at circumneutral pH. 18S rRNA gene analysis showed the low pH Mn-oxidizing fungus was closely related to Menispora tortuosa, Chaetosphaeria curvispora and Kionochaeta spissa, and the circumneutral Mn-oxidizing fungus was closely related to Myrothecium verrucaria, Didymostilbe echinofibrosa and Myrothecium roridum.

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B23G-08

Particulate Organic Carbon and Iron Speciation within Deep-Sea Hydrothermal Plumes

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Geophysical and geochemical models indicate that mid-ocean ridge hydrothermal venting may be a quantitatively significant component of the global Fe budget. Direct measurements demonstrate that seawater interactions with hydrothermal plumes exert quantitative controls on the elemental oceanic cycling of numerous trace elements and isotopes including essential nutrients and micronutrients. The chemical reactivity of hydrothermal plumes has been attributed, but not firmly linked, to the formation and surface reactivity of Fe oxide minerals. Despite the overwhelming evidence that plume particulate Fe is a key factor in ocean chemistry, essentially nothing is known of Fe speciation within plumes, outside of sulfide mineralogy. In addition, due to the analytically challenging nature of the plume particles, little is known about the mechanisms behind the biogeochemical processes catalyzed by these materials. Although a role for organic C (Corg) in Fe speciation and mobility in the deep-sea has been hypothesized, Corg sources and speciation in plumes remain largely unknown. The present study examines Fe and C speciation in plume particles at the nanometer scale using scanning transmission X- ray microscopy (STXM) combined with C 1s and Fe 2p near edge X-ray absorption fine structure (NEXAFS) spectroscopy. The results offer evidence for direct interactions between Fe and C in plume particles; Fe(II), Fe(III), and C occur in close spatial proximity within plume particle aggregates. Iron(II) is co-located with C, and is stable in the presence of dissolved O2 on timescales that exceed the calculated "half-life" (with respect to oxidation) for Fe(II) in seawater. Organic C matrices coat micrometer scale Fe-rich particles and physically entrap nanometer scale Fe-rich particles. Our results suggest a new and broader conceptual model for the source of trace-element reactive surfaces in hydrothermal plumes is required, one that includes Corg in addition to the long-hypothesized Fe oxide particles. Our study has wide ranging implications for long-range Fe oceanic transport, oceanic trace element distributions, and deep-sea and sub-seafloor microbial C fixation.

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B33A-0850

Optimization of DNA Extraction from Deep-sea Basalt

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Studies on the microorganisms that inhabit deep-sea basalt can provide information on this dark ecosystem, which will contribution to our understanding of mass transformation and energy flow in the deep ocean. However, molecular methods for use with metal- and clay-rich rock materials such as basalt have not been suitably developed at present, yet are critically required in order to be able to fully evaluate the basalt biotope. For example, inefficient DNA extraction might lead to loss of information about important components of this community, and misinterpretation about the total community diversity and function. In order to investigate the effects of sample pretreated method, particle size, different DNA extraction methods and cell density on extracted DNA yields, two basalt samples were collected from the East Pacific Rise 9° N during research cruise AT11- 20 in Nov 2004. Basalt samples were crushed to different particle size, washed with ddH_2O and 100% ethanol respectively, and autoclaved. Marinobacter aquaeolei cultures with different cell densities were inoculated into differently treated basalt samples. Pure culture and basalt samples without inoculation were used as positive and negative control to evaluate the extracting efficiency. FastDNA® spin for soil kit, GeneClean® for ancient DNA kit and UltraClean^{TM} soil DNA Kit are used for DNA extraction. Results showed that DNA yields increased with culture density. FastDNA® spin for soil kit gave the highest DNA yields, which is almost 10 times more than that of UltraCleanTM soil DNA Kit. Ethanol washing and ddH2O washing did not make big difference to DNA yields. Mineral composition and surface areas might also affect DNA yields.

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B22D-02

Anaerobic Nitrate-Dependent Metal Bio-Oxidation

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Direct biological oxidation of reduced metals (Fe(II) and U(IV)) coupled to nitrate reduction at circumneutral pH under anaerobic conditions has been recognized in several environments as well as pure culture. Several phylogentically diverse mesophilic bacteria have been described as capable of anaerobic, nitrate-dependent Fe(II) oxidation (NFOx). Our recent identification of a freshwater mesophilic, lithoautotroph, Ferrutens nitratireducens strain 2002, capable of growth through NFOx presents an opportunity to further study metal bio-oxidation. Continuing physiological studies revealed that in addition to Fe(II) oxidation, strain 2002 is capable of oxidizing U(IV) (4 µM) in washed cell suspensions with nitrate serving as the electron acceptor. Pasteurized cultures exhibited abiotic oxidation of 2 µM U(IV). Under growth conditions, strain 2002 catalyzed the oxidation of 12 µM U(IV) within a two week period. Cultures amended with sodium azide, an electron transport inhibitor, demonstrated limited oxidation (7 µM) similar to pasteurized cultures, supporting the direct role of electron transport in U(IV) bio-oxidation. The oxidation of U(IV) coupled denitrification at circumneutral pH would yield enough energy to support anaerobic microbial growth ($\Delta G°'= -460.36$ kJ/mole). It is currently unknown whether or not strain 2002 can couple this metabolism to growth. The growth of F. nitratireducens strain 2002 utilizing Fe(II) as the sole electron donor was previously demonstrated. The amount of U(IV) (~12 µM) that strain 2002 oxidized under similar autotrophic growth conditions yields 0.0019 kJ, enough energy for the generation of ATP (5.3×10^{-20} kJ ATP^{-1}), but not enough energy for cell replication as calculated for nitrate-dependent Fe(II) oxidizing conditions (0.096 kJ) assuming a similar metabolism. In addition to F. nitratireducens strain 2002, a nitrate-dependent Fe(II) oxidizing bacterium isolated from U contaminated groundwater, Diaphorobacter sp. strain TPSY, was also capable of nitrate- dependent U(IV) oxidation (8 µM over 24 hours, pseudo first order rate constant of 0.12 ± 0.02 hr-1) in washed cell suspensions. Further biochemical investigation of nitrate-dependent U(IV) oxidation in strain TPSY revealed the expression of several putative high molecular weight proteins specific to this metabolism. Together with the previously described metabolic ability of Geobacter metallireducens (Finneran et al. 2002) and Thiobacillus denitrificans (Beller 2005), these data indicate that anaerobic, metal oxidation may be a ubiquitous microbial metabolism.

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B33A-0864

Aerobic and Anaerobic Oxidation of Organic Acids in Yellowstone Hot Spring Ecosystems

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Thermodynamic analysis of energy supply based on samples collected from continental hot spring ecosystems at Yellowstone show that aerobic reactions yield the greatest energy. In terms of energy per mole of electrons transferred, aerobic oxidation of organic acids rivals or exceeds the energy supply from aerobic oxidation of hydrogen, CO, hydrogen sulfide, pyrite, sulfur or ammonia. This analysis is derived

from samples collected where hot spring fluid are in contact with the atmosphere. It is likely that oxygen will be present at lower concentrations deeper in the system, which will place hard constraints on aerobic lifestyles. If so, which metabolisms could be supported deeper in the system? How will other oxidants be used to release energy? What characterizes the transition from aerobic to anaerobic oxidation? To answer these questions, pH, temperature, and alkalinity were measured in the field while measurements of dissolved oxygen and other redox-sensitive species (nitrate, ammonia, ferrous iron, and sulfide) were made with field-portable spectrophotometers and samples were taken for analysis of organic and inorganic ions by ion chromatography. Conditions in the subsurface can be predicted by starting from measured oxygen concentrations and calculating the effect of decreasing the concentration on the overall energetics of the system. Depending on hot spring composition, the amount of energy from aerobic oxidation of organic acid anions like succinate matches that from anaerobic oxidation (by nitrate or sulfate) once the log of the activity of dissolved oxygen drops to -6 to -8. These activities are 1 to 4 orders of magnitude lower that values determined for surface water in the hot springs. At lower oxygen activities aerobic oxidation gives way to anaerobic oxidation, and organic oxidation is more likely to involve nitrate and sulfate. Preliminary estimates indicate that these changes may occur at shallow depths in hot spring sediments (perhaps within the first centimeter), which suggests great differences between conditions inferred from fluids and those inferred from genomic data based on sediments or isolates from the same hot spring system.

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B24D-03

Diversity and As-adsorption properties of Mn(II)-oxidizing bacteria within tropical wetlands of the Mekong Delta

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Manganese (Mn) and iron (Fe) oxides are ubiquitous minerals that occur under similar redox conditions in terrestrial systems and have high sorptive capacities for many trace metals, including arsenic (As). In most natural environments, Fe oxidation is dominated by abiotic processes, while Mn oxides are primarily formed via bacterial Mn(II) oxidation, and both processes can profoundly impact the mobility of metal(loid) contaminants. Deciphering the mechanisms involved in arsenic transport within soils and sediments is essential for aiding many Southeast Asian countries, including Cambodia, where naturally occurring As is in groundwater at concentrations well above the WHO recommended limit. Although numerous past studies have characterized the effects of As adsorption onto Fe and Mn oxides individually, it is unknown whether, in the presence of both oxides, there is preferential adsorption of As onto one oxide over the other. In the present study, we examine the competitive retention of As(III) and As(V) on goethite and biogenic Mn oxides using Donnan membranes--where each oxides is isolated by a semi-permeable membrane through which arsenic can migrate. Mn(II)-oxidizing bacteria, isolated from several Mn-rich sites along the Mekong River and wetland areas within the Mekong delta, were dominated by a diverse array of Bacillus strains that rapidly oxidize Mn(II) within three to five days in liquid culture. The results of this study not only expand our knowledge of the diversity and biogeochemical importance of terrestrial Mn(II)-oxidizing bacteria, but also contribute to our understanding the relative impact of Fe and Mn oxides on arsenic retention within natural wetlands.

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