

harsh Antarctic environment is limited, because traditional methods of studying microbial physiology are very difficult to apply to organisms with extremely low levels of metabolic activity. In order to fully understand carbon and nitrogen cycling and nutrient uptake in cryptoendolithic communities, and the metabolic costs that the organisms incur in order to survive, it is necessary to employ molecular geochemical techniques such as amino acid analysis in addition to physiological methods. Low-molecular-weight biomolecules such as amino acids can be used as tracers of carbon and nitrogen uptake and loss by microbial communities living in solid-state matrices such as rock or sediment.

We have measured the concentrations and D/L ratios for several amino acids as a function of depth in a large sandstone boulder. Concentrations of both free and bound amino acids decrease by more than two orders of magnitude from the surface to the visible base of the community (approximately 1.2 cm depth), while the D/L ratios of the amino acids increase from near zero to 0.2 or greater over the same depth interval. We interpret these data as an indication that one or more community members are selectively scavenging L-amino acids as the amino acids are transported through the rock by intermittently percolating meltwater. This is consistent with the known preference of lichens for amino acids as nitrogen sources rather than inorganic nitrogen under conditions of nutrient limitation. It is not yet clear whether there is also a contribution to amino acid uptake from heterotrophic bacteria associated with the cryptoendolithic community. The increase in D/L ratios with depth observed in the rock is too great to be attributable solely to the natural occurrence of D-amino acids in bacteria.

Amino acid concentration and D/L profiles remain relatively constant below the 1.2 cm level. This may be due to aqueous transport from the upper levels. It is also possible, however, that heterotrophs at very low cell densities may exist several cm below the bottom of the bulk endolithic community.

B11A-0721 0830h POSTER

Amino acid and hexosamine in the equatorial western Pacific: vertical fluxes and individual preservation through water column to surface sediments

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Amino acids (AA) and hexosamines (HA) are major constituents for all living organisms, constituting important fractions of labile organic carbon and nitrogen. They usually decompose rapidly than bulk OM and must be expected to be closely linked to biogeochemical processes. In spite of such importance, our understanding of degradation processes of labile components is still limited. Therefore vertical fluxes and preservation of AA and HA from water column to surface sediments are investigated at the western equatorial Pacific. The settling particles were composed of fairly fresh AA, which could be derived from siliceous diatom with less amount of calcareous plankton. In contrast, AA were degraded in sediments and porewaters. Each AA showed highly variable preservation ratio from settling to sedimentary particles. Compared with glycine, the calculated preservation ratio was the lowest (0%) for cysteine, followed by phenylalanine (6%), tyrosine (17%), methionine (47%), leucine (60%), isoleucine (65%), proline (67%), valine (91%), serine (99%), arginine (107%), threonine (112%), alanine (115%), glutamic acid (114%), aspartic acid (150%), lysine (166%) and histidine (186%). Beta-alanine and gamma-aminobutyric acid were the least labile AA. Probably they are so difficult to degrade for bacteria to get biochemical energy that the degradation proceeds fairly slowly. In contrast, after burial, even most labile, aromatic and sulfur-containing AA, degrade at a rate similar to the other protein AA. In spite of complicated reactions, most of the AA showed first-order reaction kinetics during the degradation in the sediments. The decomposition rate constant k (kyr⁻¹) in this study was 2-3 orders lower than those in coastal marine environments. Better preservation of HA over AA in the sediments was probably due to the general incorporation of HA into structural biopolymer matrices, such as bacterial cell-walls and chitinous material. Abundant glycine in the AA in the sediments is due to contribution from diatom cell-walls, bacterial peptidoglycan, and the degradation by bacterial activity. Dissolved combined AA (DCAA) showed enrichment in glutamic acid, glycine and threonine, and depletion in aspartic acid and alanine. Bacterial biomass and/or

activity influences DCAA in porewaters more than AA in the sediments. Phenylalanine was abundant in the dissolved free AA (DFAA). Both aromatic and acidic AA are generally concentrated in diatom cell protoplasm, which is more rapidly degraded than cell-walls. Good correlation between aspartic acid and carbonate contents in the sediments and poor correlation in the settling particles indicates that aspartic acid is significantly controlled by the reaction or adsorption with carbonates during early diagenesis. Abundant occurrence of clay minerals in sediments would be responsible for the enhanced accumulation of basic AA and arginine. During diagenesis, bulk Corganic/N ratios are mainly controlled by more contribution of ammonium, which is incorporated into the lattice of clay minerals, not by the compositional change in AA. Microbial degradation continued to reduce AA and OM in the sediments, which has implications for appreciable under-estimates of paleoproductivity.

B11B MCC: Hall C Monday 0830h

Aqueous Microbial Geochemistry: Extreme and Contaminated Environments I Posters (joint with H, OS)

Presiding: M E Kauffman, Idaho National Engineering and Environmental Geomicrobiology Group; L A Warren, McMaster University

B11B-0722 0830h POSTER

The Reductive Immobilization of Pertechnetate by Bioreduced Sediments

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Sediments from the Hanford Site, WA, and Oak Ridge, TN, were incubated for up to 60 d with *Shewanella putrefaciens*, washed and pasteurized, then contacted with 20 micromolar pertechnetate. The clastic Hanford sediment was fluvioacustrine in origin, and the Oak Ridge sediment was fine weathered shale. In Hanford sediment only 1% of the total Fe (8 wt. %) was reduced after 50 d incubation; in Oak Ridge sediment 18% of the total Fe (5 wt. %) was bioreduced, 72% of the Hanford Mn (1 wt. %), and all of the Oak Ridge Mn (0.2 wt. %) were bioreduced. Mn(III/IV) oxides buffered the sediment redox potential and inhibited Tc(VII) reduction. Examination by EMP and XMP showed the absence of discrete Mn(II) solids after bioreduction. Individual microXANES analyses of Hanford sediment indicated the presence of Mn(II) and Mn(III/IV), even after Mn bioreduction reached a terminal state after 23 d. In Hanford sediment Tc(IV) was associated with Fe-Mn oxides and weathered biotites including interlamellar Fe-Mn oxides. On the biotites, Tc(IV) was concentrated at grain boundaries, where Fe and Mn were most available to microbial reduction. In Oak Ridge sediment, Tc(IV) was associated with one of two morphotypes of weathered shale.

B11B-0723 0830h POSTER

Metal Reduction and Mineral formation by a Psychrotolerant Fe(III)-Reducing Bacterium Isolated from an Iron-Rich Waters near a Hydrothermal Vent

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Although dissimilatory metal reduction and mineral formation under mesophilic and thermophilic conditions are extensively examined, they are poorly understood under low temperature. The objective of this study was to examine metal reduction and mineral

formation using a psychrotolerant iron-reducing bacterium (*Shewanella alga*, PV-4) isolated from iron-rich waters associated with the Naha vents off the Hawaiian coast. The psychrotolerant iron-reducing bacterium was able to use lactate, formate, and hydrogen as an electron donor while reducing Fe(III)-citrate, Fe(III)-EDTA, Co(III)-EDTA, Cr(VI), Mn(IV), and iron oxyhydroxide (FeOOH) at temperatures between 0 and 37°C. The psychrotolerant bacterium exhibited diverse mineral precipitation capabilities including the formation of magnetite (Fe₃O₄), siderite (FeCO₃), and rhodochrosite (MnCO₃). Transmission electron microscopic data showed that PV-4 formed mainly superparamagnetic magnetite at temperatures ranging from 0 to 14°C and formed mainly single-domain magnetite at temperatures ranging from 18 to 37°C. This study indicates that iron-reducing bacteria may contribute to the biogeochemical cycling of metals and carbon at low temperatures and may contribute to the natural remnant magnetism of marine sediments.

B11B-0724 0830h POSTER

Metal Reduction and Mineral Formation by an Alkaliphilic Fe(III)-Reducing Bacterium Isolated from an Alkaline Leachate Pond

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Microbial metal reduction plays an important role in biogeochemical cycling of carbon and has the potential for immobilizing metals and radionuclides in diverse environments. The objective of this study was to examine metal reduction and mineral formation using an alkaliphilic bacterium, *Alkaliphilus* (QYMF), isolated from a leachate-pond containing high levels of salt (Na concentration = 440 - 12,100 ppm) and boron (2,000 - 3,000 ppm) at pH 9.0-10.0. The bacterium was able to use lactate, acetate and hydrogen as alternative electron donors and Fe(III)-citrate, Fe(III)-EDTA, selenate, Cr(VI), Co(III)-EDTA, and iron oxyhydroxide (FeOOH) as electron acceptors. The reduction of Fe(III)-citrate and Fe(III)-EDTA in the presence of H₂PO₄ and boron resulted in the precipitation of vivianite [Fe₃(PO₄)₂·8H₂O]. Formation of sparingly soluble precipitates, mediated by the alkaliphilic Fe(III)-reducing bacterium, may sequester iron, phosphate, and other metals into more stable and less toxic forms. These results suggest that bioremediation of metal-contaminated alkaline environments may be feasible, and that the process of metal-reduction may occur in alkaline habitats.

B11B-0725 0830h POSTER

Effect of Sediment Mineralogy on Microbiologically Induced (DMRB) Changes in Divalent Metal Speciation

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Dissimilatory metal reducing bacteria (DMRB) can either directly mediate or indirectly induce geochemical processes that alter the speciation and lability of metallic contaminants within natural environments. Most investigations into the effect of DMRB on metal speciation utilize synthetic iron oxyhydroxide minerals as the Fe(III) source, thereby allowing well-controlled experiments. However, this technique does not emulate the actual mineralogical composition of natural systems and does not account for the small-scale heterogeneity that may control metal geochemistry within these systems. Our experiments with a divalent metal that is subject to both surface complexation and ion exchange reactions (Zn²⁺) indicate that clay minerals place an important control on DMRB-induced changes in metal speciation. Our data demonstrates that microbial Fe(III) reduction alters the proportion of Zn-aq

in slurries containing goethite and hydrous ferric oxide (HFO), but has little effect in parallel slurries containing selected iron-bearing natural sediments. These data also indicate that microbial goethite and HFO reduction increases the proportion of HCl-insoluble Zn while microbial reduction of Fe(III) in natural sediments has virtually no effect on HCl-insoluble Zn. Geochemical modelling indicates that Zn may be released from surface complexes (via Fe-reduction) and then react with ion exchange sites on clay mineral surfaces in natural sediments. This process can prevent net solubilization of Zn, and prevent Zn from participating in the surface reactions that occur in the corollary experiments with pure iron oxyhydroxide minerals. The Zn-speciation changes in the goethite system have been previously reported by these investigators, and may indicate that DMRB are inducing processes that enhance iron-oxyhydroxide recrystallization rates thus incorporating Zn into less soluble iron oxide minerals. Mssbauer analyses support this hypothesis. XAS data indicate the Zn-O bonding environment is initially similar in both sediments. However, the Zn-O co-ordination number shifts during natural sediment reduction but remains constant during goethite reduction. Although alternate explanations are possible, this shift could represent Zn changing from an iron-oxide surface complex to an ion exchange site. Scanning surface mass spectroscopy analyses of elemental abundance associated with adsorbed Zn in these systems are underway and will be discussed. These results indicate that metal mobility predictions based on microbial experiments with synthetic Fe-oxide minerals may have limited applicability to real world systems.

B11B-0726 0830h POSTER

Gram-Positive Nickel Resistant Bacteria Isolated from Riparian Sediments Contaminated with Ni and U on the Savannah River Site

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The natural attenuation of pollutants in riparian and wetland systems is driven in large part by the services provided by the diverse microbial communities that thrive in these nutritionally and chemically complex environments. For co-contaminated systems, the presence of heavy metals at excessive levels may alter the structure and function of microbial communities that are essential for the immobilization of inorganics and degradation of organic contaminants. We examined riparian sediments heavily contaminated with U and Ni (1000s of mg/kg) from a small stream on the U.S. Department of Energy's Savannah River Site that received metallurgical process effluents wastewater over a thirty-year period associated with the production of nuclear materials. Four gram positive bacteria were isolated that displayed marked resistance (5000 mg/kg) to Ni relative to organisms from uncontaminated control locations: *Arthrobacter oxydans*, *Streptomyces galbus*, *Streptomyces aureofaciens*, and *Kitasatospora cystarginea*. The metal resistance of *S. aureofaciens* and *K. cystarginea* was further characterized in growth experiments for resistance to other metals. Ongoing geochemical characterization of U and Ni in terms of solid phase partitioning and aqueous phase speciation and solubility indicate that Ni is more chemically labile and, by extension, bioavailable than U in these aged-contaminated sediments. Accordingly, the isolation of Ni resistant organisms is consistent with greater selective pressure from Ni as a result of its greater bioavailability. These results are placed in context of environmental management and remediation of co-contaminated, biogeochemically complex environments.

B11B-0727 0830h POSTER

Kinetics of Calcite Precipitation Induced by Ureolytic Bacteria at 10 to 20 °C in Artificial Groundwater

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The ureolytic bacterium *Bacillus pasteurii* can induce calcite precipitation through the metabolic hydrolysis of urea. Metabolic urea hydrolysis results in the production of ammonium and carbonate ions and an increase in pH, which can induce the precipitation of calcite. Because calcite precipitation may result in solid phase capture of contaminant metals, the bacterially mediated precipitation of calcite is of particular interest for bioremediation purposes.

In this study, the kinetics of calcite precipitation induced by bacterially mediated hydrolysis of urea is investigated. Experiments were performed in microcosms inoculated with *Bacillus pasteurii* ATCC 11859 and containing an artificial groundwater prepared to mimic the aqueous chemistry of the Snake River Plain Aquifer, Idaho Falls, U.S.A. Microcosms also contained 6.0 mM urea (for experiments ran at 20 °C) and 25 mM urea (for experiments ran at 10 and 15 °C). Throughout the experiment the experimental solutions were monitored for ammonium, dissolved magnesium, dissolved calcium content and pH. Experimental data was fit to integrated equations for urea hydrolysis, calcite saturation state and dissolved calcium concentrations using unconstrained nonlinear regression and a quasi-Newton optimization routine for parameter estimation (STATISTICA v 5.0).

Rates of urea hydrolysis showed a pronounced inverse temperature dependence with rate constants (k_{urea}) of 0.09, 0.18 and 0.91 at 10, 15 and 20 °C respectively. The production of ammonium and carbonate from urea hydrolysis increased the pH at all temperatures to 9.3 in less than one day. Coupled with this, dissolved calcium concentrations started to decrease after one day at 20 and 15 °C and 2 days at 10 °C. These decreases in dissolved calcium were accompanied by the precipitation of calcite.

Although dissolved Ca^{2+} depletion over time did not appear to correlate directly with the rates of urea hydrolysis, a comparison of Ca^{2+} depletion with ammonium production reveals calcite precipitation was initiated at similar (approx. 8.0 mM) ammonium concentrations at all temperatures. This indicates calcite precipitation was triggered by approximately the same degree of oversaturation at all temperatures.

As calcite precipitation occurred, calcite saturation state decreased. Interestingly, rate constants (k_s) for the change in saturation state were similar at all temperatures (0.27, 0.25 and 0.26 for 10, 15 and 20 °C respectively). This is to be expected considering calcite saturation state calculations included the effect of temperature. Furthermore, rate constants for calcite precipitation k_p and the critical saturation state required to cause precipitation ($S_{critical}$) also did not vary notably with temperature.

Interestingly, the rates of calcite precipitation observed here are one to two orders of magnitude lower than those observed in other studies. This can be explained by the presence of Mg^{2+} (in the artificial groundwater) and NH_4^+ (produced by bacterial urea hydrolysis) which may adsorb onto and mask active nucleation sites. Thus *B. pasteurii* may induce calcite precipitation through urea metabolism, but may also slow down calcite precipitation kinetics through the production of ammonium. Clearly the role of bacterially mediated urea hydrolysis in calcite precipitation is a complex one which requires significant further study for use in bioremediation.

B11B-0728 0830h POSTER

Strontium Incorporation Into Calcite Generated by Bacterial Ureolysis

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Strontium incorporation into calcite generated by bacterial ureolytic activity was investigated as part of a larger effort to evaluate the use of *in situ* urea hydrolysis for accelerating co-precipitation of trace metals and radionuclides in contaminated aquifers. ⁹⁰Sr, a uranium fission product with a half-life of 29 years, is a significant subsurface contaminant at several Department of Energy facilities and could be immobilized using this remediation strategy. Experiments were conducted in a medium designed to simulate the groundwater of the Snake River Plain Aquifer in eastern Idaho, amended with strontium. Initially the solution was undersaturated with respect to calcite. As a model ureolytic organism, we used *Bacillus pasteurii*, a well-characterized

bacterium known for high urease activity and previously shown to induce calcite precipitation in urea-amended medium. To gain information on the effect of the bacterial surfaces, we also looked at precipitation in the presence of a bacterial species that did not hydrolyze urea, as well as in the absence of bacteria. In the absence of bacterial ureolysis, carbonate precipitation was induced by addition of ammonium carbonate. All products were identified as calcite by X-ray diffraction. Strontium uptake was observed in all cases, but was greatest in the system including bacterial ureolysis. Sputter depth element profiling by time-of-flight secondary ion mass spectrometry (TOF-SIMS) confirmed this finding, showing highest Sr:Ca ratios in the bacterially generated calcite throughout the depth (350 nm) investigated. Environmental Scanning Electron Microscopy (ESEM) of the solids revealed regular crystals containing the outlines of embedded or entombed bacterial cells, suggesting that calcite precipitated directly on the cell surfaces when present. Analysis by X-ray Absorption Near Edge Spectroscopy (XANES) indicated that in both the biotically and abiotically generated calcites the Sr was incorporated into the calcite lattice structure, rather than forming strontium carbonate. These findings are encouraging with respect to long term containment and stabilization of strontium by the proposed remediation strategy.

B11B-0729 0830h POSTER

The Biogeochemistry of Phosphate Mineral Dissolution

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It has been found that different microorganisms dissolve phosphate minerals in soil to supply needed phosphorus. The growth of various bacteria and fungi, when not supplied with necessary phosphorus in lab tests, have shown some of these microorganisms to secrete weak acids, dissolving phosphate minerals to various degrees. This study attempted to evaluate such solubilities.

Aspergillum niger, a common fungi, was grown in a liquid media for a two day period, filtered, and then added to finely ground samples of the phosphate minerals apatite, variscite, and lazulite. The filtrates were tested for pH and phosphate concentrations before and after addition of the minerals.

Initial results indicate pH of the filtrates has a direct correlation with phosphate concentrations. The different phosphate minerals showed different solubilities in the filtrates. Collophane, an amorphous variant of apatite also tested, showed a significantly greater phosphate concentration in the filtrate than the crystalline apatite sample.

B11B-0730 0830h POSTER

Biogeochemical Cycling of Sulfur in Soil

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Sulfur is an important element of the global biogeochemical cycle, since it is highly reactive and moves freely among the lithosphere, atmosphere and hydrosphere. Climatic and environmental changes affecting sulfur in the pedosphere will inevitably change the rate and forms of global sulfur cycling which are intertwined with that of carbon, nitrogen and phosphorus. In soil, inorganic sulfur derived from atmospheric deposition or fertilization is largely immobilized and incorporated into soil organic matter (>95%). During the last decades, however, these emissions have

been significantly reduced in North America and Europe, and S deficiency can increasingly be observed in crops. This process was accelerated by a change to low-S-containing fertilizers. Therefore, we studied the long-term dynamics of S forms in relation to organic C to evaluate its impact on the soil cycle. Synchrotron-based sulfur K-edge X-ray Absorption Near-Edge Structure spectroscopy (XANES) was used to speciate and quantify the different oxidation states of soil sulfur (organic and inorganic forms of S). Direct measurement of S species in bulk soil indicated the presence of large background on the spectra, which could not easily be corrected without affecting the results. However, humic acid extractions using 0.1 M NaOH/0.4 M NaF mixtures produced better signals, which can even be improved by additional filtration using a 0.2mm membrane filter under pressure. Traditional wet chemical analyses of soil S using hydriodic acid (HI) reduction showed that the major proportion (98%) of total S was present in organic forms, out of which 77-84% were C-bonded S, whereas ester SO₄-S constituted merely 16-23% of the organic S pool in bulk soils. These values were constant regardless of major soil disturbances by landuse and did not change between different particle size fractions. S-XANES spectroscopy, however, showed clear differences of S oxidation states after environmental disturbance of soil and between physical soil separates making it a useful tool to assess S cycling in soil.

B11B-0731 0830h POSTER

Elemental Sulfur Reactions at Low pH and Their Implication for Microbial Activity in Acid Mine Drainage Environments

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Elemental sulfur oxidation in low pH, oxic conditions is a key intermediate process governing the oxidation and acid generating potential of metal sulfide minerals. Nucleophilic reactions with aqueous sulfur intermediates, reactions with intermediate oxygen species, and oxidation reactions with ferric iron and/or oxygen are all important in considering how microorganisms utilize elemental sulfur for energy. A series of experiments investigating the kinetics of these reactions illustrates some of the requirements for sulfur-oxidizing microorganisms to utilize elemental sulfur in low pH environments.

Surface area-normalized rates for the oxidation of elemental sulfur with molecular oxygen are on the order of 5 x 10⁻¹⁰ mol m⁻² sec⁻¹ at pH 1.5 and 1 x 10⁻⁸ mol m⁻² sec⁻¹ at pH 0.5. Ferric iron accelerates the rate by approximately a factor of 2, but this rate is independent of the ferric iron concentration over 2 orders of magnitude, within experimental error. Reactivity of elemental sulfur is not markedly different whether the sulfur is in the mineral structure or as a free S₈ ring. Flocculation of small particles of elemental sulfur may severely limit the reactivity of this mineral in inorganic experiments. Surface infrared spectroscopy measurements did not show the development of any resolvable sulfur-oxygen bonds in 20 hours of reaction with 1 mM Fe³⁺, H₂O₂, or OH* (as Fentons reagent) at pH 2.

Observations of rate dependences from this study suggest that microbial utilization of elemental sulfur may be limited by a reaction involving the S₈ ring, and not single S₀ atoms in solution. The extremely low solubility of elemental sulfur in water and the tendency of small elemental sulfur grains to flocculate illustrate the potential importance of extracellular material as either a surfactant to import an S₈ ring inside the cell or to facilitate a reaction to transform S₈ into a more soluble and easily transported species. Changes in nucleophilicity of previously identified enzymes at low pH indicate that they would not function at low pH and therefore extracellular utilization of elemental sulfur must proceed by a different mechanism under these conditions. We will present some of the geochemical requirements for enzymatic function in low pH environments and report on current work towards identifying and characterizing enzymatic compounds that facilitate microbial functions in these environments.

B11B-0732 0830h POSTER

Reactivity of the Bacteria-Water Interface: Linking Nutrient Availability to Bacteria-Metal Interactions

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Identifying and quantifying the controls on metal mobilities in geologic systems is critical in order to understand processes such as global element cycling, metal transport in near-surface water-rock systems, sedimentary diagenesis, and mineral formation. Bacteria are ubiquitous in near-surface water-rock systems, and numerous laboratory and field studies have demonstrated that bacteria can facilitate the formation and dissolution of minerals, and enhance or inhibit contaminant transport. However, despite the growing evidence that bacteria play a key role in many geologic processes in low temperature systems, our understanding of the influence of the local nutrient dynamics of the system of interest on bacteria-metal interactions is limited. Here we present data demonstrating the effectiveness of coupling laboratory experiments with geochemical modeling to isolate the effect of nutrient availability on bacterially mediated proton and metal adsorption reactions.

Experimental studies of metal-bacteria interactions were conducted in batch reactors as a function of pH, and solid-solute interactions after growth in a variety of defined and undefined media. Media nutrient composition (C,N,P) was quantified before and after harvesting the cells. Surface complexation models (SCM) for the adsorption reactions were developed by combining sorption data with the results of acid-base titrations, and in some cases zeta potential titrations of the bacterial surface. Our results indicate a clear change in both buffering potential and metal binding capacity of the cell walls of *Bacillus subtilis* as a function of initial media conditions. Combining current studies with our past studies on the effects of growth phase and others work on temperature dependence on metal adsorption we hope to develop a holistic surface complexation model for quantifying bacterial effects on metal mass transfer in many geologic systems.

B11B-0733 0830h POSTER

Ternary Complexation on Bacterial Surfaces: Implications for Subsurface Anion Transport

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The physical, chemical, and biological controls on contaminant mobilities in aquatic ecosystems must be determined to establish the threat that contamination poses to the environment. Quantitative models of contaminant mobilities are required as a prerequisite to guide remediation efforts and to prioritize the potential hazard to the ecosystem of each contaminated site. It is well established that mineral surface adsorption is an important control on contaminant mobilities, and many studies have utilized thermodynamics to quantify metal/organic adsorption in order to yield predictive models of contaminant transport. However, these models of contaminant transport may not be representative of the reactions which control contaminant mobilities as most mineral surfaces are coated with organic acids, bacteria, and extracellular polymers. Numerous laboratory studies have demonstrated that bacterial cell walls have a high affinity for binding metal cations, and field studies indicate that a significant proportion of bacteria cells and associated extracellular matrices are coated with small scale hydrous metal oxides. The small size of bacteria, and in many cases the nanoscale of their associated mineral phases, suggests these bacteria-mineral composites may represent a large proportion of surface area exposed to fluid flow. Therefore, due to the affinity of bacterial cell walls for cations and biomaterials, bacteria may also have a significant impact on anionic contaminant mobility in many natural systems.

The extent of metal-bacteria adsorption reactions varies drastically as a function of pH and solution chemistry. Current adsorption models have focused on the interactions of positively charged metal cations

with bacterial surfaces, however in many oxidizing environments metals such as Cr exist as anions or anionic complexes. We have studied the ability of non-metabolizing cells of the bacterial species *Bacillus subtilis* and *Shewanella putrefaciens* to adsorb aqueous Cr(VI) and I⁻ in the presence of background electrolyte and aqueous Al (III), Cd(II), Ca(II) or cells coated with Al (oxy)hydroxide phases. We use a unique blend of XRD, electrophoretic mobility, SEM, and aqueous geochemistry measurements to quantify the mechanisms of Cr(VI) and I⁻ removal from solution. Our results indicate the removal of both anions is highly dependent on solution pH with significant removal at low pH and diminishing removal at higher pH values, without the presence of cations or precipitates. However, in the presence of aqueous Cd(II) and Ca(II) which adsorb strongly from pH 3.5-8, the removal of Cr(VI) and I⁻ increases appreciably. Furthermore the loading of the cell surface with small amorphous mineral phases increases adsorption. Aging of the mineral-bacteria composites appears to decrease removal efficiency due to coarsening of the mineral phases and a decrease in charge density. Considering that many geologic environments include both cationic and anionic metal contaminants, our results suggest that mass transport of Cr(VI), I⁻, and other anions may be affected by ternary complexation or other cation mediated surface reactions in bacteria-bearing systems.

B11B-0734 0830h POSTER

Adsorption of Ferrous Ions onto *Bacillus subtilis* Cells.

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Secondary iron-oxides forming by the oxidation of ferrous ions have been observed to accumulate on the surface of bacterial cells in various environments (e.g. lake sediments). It is not always clear to which extent the presence of the bacteria can affect the precipitation process. The cells may play an active (metabolic) role in the oxidation of the ferrous ions. Whether this is the case or not, it does not change the fact that bacterial surfaces can also sorb ferrous/ferric iron and crystal nuclei and therefore influence the properties of the Fe-rich crystals. In order to determine the relative roles of active and passive processes, an important step is to quantify the interactions between ferrous ions and bacterial surfaces.

We have measured the kinetics of adsorption and the pH-adsorption/desorption isotherms of ferrous ions on the surface of *Bacillus subtilis* cells under anaerobic conditions, using various sorbent/sorbate ratios. In conjunction with pH-titrations, this allowed us to estimate the adsorption constants of the ferrous ions onto the various chemical sites that are present on the surface of the cells (e.g. -log(K) 3-45 for the carboxylic groups). We find that the adsorption of the ferrous ions is a quick but reversible process, which can be modeled by statistical thermodynamics. We will describe our results and discuss their consequences in the example of a simple experience of biomineralization realized in the laboratory with *Bacillus subtilis* cells.

B11B-0735 0830h POSTER

The Impact of Disturbance of Biological Soil Crusts on Soil Nitrogen Content and Isotope Composition, and Potential Microbial Respiration and Nitrogen Mineralization in Four Arid Ecosystems

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Biological soil crusts are widespread in arid ecosystems and can be the primary source of new nitrogen inputs into soils. Disturbance of these crusts can have

long-term consequences on plant and soil carbon and nitrogen dynamics. We experimentally disturbed soil crusts in Great Basin, Colorado Plateau, Chihuahuan, and Sonoran Desert ecosystems to quantify the immediate effects (one to two years) of disturbance. The nitrogen isotope composition of soil crusts indicates that nitrogen fixation is the primary source of nitrogen at sites on the Colorado Plateau and in the Sonoran Desert, but not at sites in the Great Basin or Chihuahuan Desert. Disturbance did not significantly change soil nitrogen content at any site. Soil nitrogen isotope composition did increase 1.5 per mil following disturbance in the Sonoran Desert, but did not significantly change at other sites. Disturbance decreased potential microbial respiration by 25% on the Colorado Plateau, but increased potential nitrogen mineralization in the Sonoran and Chihuahuan Deserts. The short-term consequences of disturbance are site-specific so scientists should avoid broad generalizations on the short-term impact of disturbance in arid ecosystems.

B11B-0736 0830h POSTER

Net N and C mineralization and soil microbial biomass with elevated CO₂ in a Mojave Desert ecosystem

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We quantified changes in soil microbial activity and N availability with elevated CO₂ in a Mojave Desert ecosystem by measuring microbial biomass N and net N and C mineralization in long-term (280 d) incubations of soil from four different cover types: *Lycium* spp., *Larrea tridentata*, *Pleuraphis rigida*, and plant interspaces. We also measured soil respiration in the field under the same cover types. There was no effect of elevated CO₂ on microbial biomass N. Soils from under *Larrea* and *Lycium* had higher microbial biomass N (52.94 mg kg⁻¹) than soils from plant interspaces (31.55 mg kg⁻¹). Soil δ¹⁵N increased during the incubation (7.3 vs. 6.3‰), reflecting microbial activity; but there was no difference in soil δ¹⁵N with CO₂ treatment. Carbon mineralization for soils under *Lycium* was 40% greater with elevated CO₂ for the last four of the seven extraction dates. Soils from interspaces had significantly lower amounts of C mineralized (1.89 ± 0.11 g kg⁻¹) than soils from under shrubs (2.99 ± 0.38 g kg⁻¹). On the last extraction date, soils from under *Lycium* experienced 40% greater net N mineralization with elevated CO₂. Soils from under shrubs experienced greater cumulative net N mineralization (102.64 ± 24.15 mg kg⁻¹) than soils from under grasses and plant interspaces (39.96 ± 9.69 mg kg⁻¹). Mean soil respiration across all cover types was higher with elevated CO₂ (2.72 ± 0.50 vs. 1.59 ± 0.29 μg C m⁻² s⁻¹). These results indicate four findings: 1) microbial activity can result in shifts in bulk soil δ¹⁵N; 2) shifts in soil microbial activity and/or structure may occur with elevated CO₂; 3) these shifts may increase gross N mineralization, but increases in microbial immobilization can limit changes in net mineralization; and 4) increased rhizosphere activity can occur with elevated CO₂. We suggest that elevated CO₂ may induce increases in rhizodeposition and/or soil microbial activity that promote both gross N mineralization and immobilization.

B11B-0737 0830h POSTER

Microbial Communities and a Novel Symbiotic Interaction in Extremely Acidic Mine Drainage at Iron Mountain, California

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Culture-independent studies of microbial communities in the acid mine drainage (AMD) system associated with the Richmond ore body at Iron Mountain, CA, demonstrated that the total number of prokaryote lineages is small compared to other environments. Phylogenetic analyses of 232 small subunit ribosomal RNA (rRNA) genes from six clone libraries revealed some novel lines of descent. Many of the novel clones were from libraries constructed from subaerial biofilms associated with fine grained pyrite. The clones form several distinct groups within the order Thermoplasmatales and are most closely related to *Ferroplasma* spp. and *Thermoplasma* spp. Another novel group

detected in a pH 1.4 pool and a pH 0.8 biofilm falls within the Rickettsiales (alpha-proteobacteria and related to mitochondria) and is most closely related to alpha-proteobacterial endosymbionts of *Acanthamoeba* spp. An oligonucleotide rRNA probe designed to target alpha-proteobacteria revealed that these are protist endosymbionts, and that they are associated with a small percentage (2%) of the total eukaryotes in samples from the Richmond mine. Measurements of the internal pH of these protists show that their cytosol is close to neutral. Thus, protists provide a habitat within the AMD system that is at least 5 pH units less acidic than the surroundings. The uncultured AMD endosymbionts have a conserved 273 nucleotide intervening sequence (IVS) in the variable V1 region of their 16S rRNA gene. The IVS does not match any sequence in current databases, but predicted secondary structure form well defined stem loops. The discovery of inserts within a highly conserved gene is extremely rare. At present we have not identified the protist host. However, it is interesting to note that protists previously shown to have alpha-proteobacterial endosymbionts possess 18S rRNA genes that contain both IVSs and group I introns. The possibility that the IVS in the AMD bacteria is a result of extensive genetic exchange between a symbiont and its host is discussed. This is the first report of a prokaryotic/eukaryotic symbiosis in an AMD microbial community. These endosymbionts add to the metabolic repertoire and provide pathways for lateral gene transfer between neutrophilic and acidophilic lineages.

B11B-0738 0830h POSTER

Discovery of an Active Microbial Community in a Subglacial Volcanic Crater Lake, Iceland

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Grímsvötn, an active volcano beneath the Vatnajökull glacier in Iceland, hosts a subglacial caldera lake (Gudmundsson, Sigurdsson & Björnsson 1997 Nature 389, 954). Except for earlier geochemical measurements (Ágústsdóttir & Brantley 1994 J. Geophys. Res. 99, 9505), the lake and its possible biota have remained unexplored. In June 2002 we penetrated the 300-meter ice sheet over the lake using hot-water drilling and collected water and tephra sediment samples. The 85°C drilling water and chemical sterilization of equipment were used to minimize sample contamination. Samples of borehole water, glacial ice, and snow were also obtained. Lake water was at the freezing point and samples had no sulfidic smell indicative of anaerobic conditions. One sample from the borehole actively degassed after retrieval. Waters were slightly acidic (pH ≈ 5) and fresh ([Na] = 5 ppm) with low sulfate (2 ppm). Elevated transition metal levels measured by ICP-MS are being analyzed. Direct (DAPI stain) cell counts in water and sediment average 2 × 10⁴ cells ml⁻¹ and 4 × 10⁷ cells ml⁻¹, respectively. Counts on R2A plates incubated under aerobic conditions at 22°C and 6°C for one week were 1 – 2 × 10⁴ and 5 × 10³ CFU/ml, respectively. These values may reflect growth during sample shipment, however, growth at 6°C indicates the presence of psychrophilic or at least psychrotolerant organisms in the lake. Colonies from lake and post-penetration borehole samples were distinct from those of the borehole, ice and snow. Incubations of lake samples at 4°C showed incorporation of ¹⁴C (from bicarbonate). Incubations of other aliquots with a nitrogen-acetylene mixture indicated production of ethylene suggestive of nitrogenase activity. DNA will be extracted and analyzed to determine if the microbial community is distinct from that in the overlying snow and ice.

B11B-0739 0830h POSTER

Subglacial Microbial Communities and Their Relationship to Bedrock Lithology

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Recent studies have demonstrated the presence of abundant and metabolically active microbial communities in subglacial systems. However, the composition of these communities and the effect that they have on the geochemical dynamics of these systems is unclear. We compared the microbial community composition of samples from two glaciers: the Bench Glacier, Alaska, USA (BG), which overlies Cretaceous metasediments, and the John Evans Glacier, Ellesmere Island, Nunavut, Canada, (JEG) which overlies a carbonate/evaporite sequence of Cambrian/Ordovician/Silurian age. Bulk meltwater chemistry indicates that sulfide oxidation and carbonate dissolution account for 90% of the solute flux from BG, whereas gypsum/anhydrite and carbonate dissolution alone accounts for most of the flux from JEG. Microbes are abundant in the subglacial environment (ca. 1.5 × 10⁵ cells ml⁻¹) at both sites based on DAPI direct counts. Furthermore, early season subglacial waters (2002) at BG, have higher counts than both supraglacial and ice marginal environments. These data are consistent with previous findings in other subglacial systems.

Two hundred and thirty three 16S rDNA clones were recovered from subglacial samples from the two glaciers in 1999 and grouped by RFLP analysis, followed by phylogenetic analysis of gene sequences. Interestingly, many (46%) 16S rDNAs from BG were most closely related to organisms that are capable of oxidation of sulfide and/or iron, whereas only a few (7%) of rDNAs from JEG were related to similar groups. The abundance of putative sulfide oxidizers in BG and their scarcity in JEG correlates with the meltwater chemistry of the two systems. These results are consistent with the hypothesis that microbial activity has an important role in contributing to the solute flux from glaciers.

Group-specific phylogenetic probes have been developed for rDNAs that are abundant in these clone libraries. These probes are currently being used for quantitative dot-blot hybridization on samples taken in 2001/2002 from a range of environments at the two glaciers to assess a) if the microbial populations are stable through time and b) the spatial variability of populations at each glacier. The studies reported here are the first to examine the composition of microbial communities in contrasting glaciated catchments using molecular techniques. The data imply a microbial influence on subglacial geochemical dynamics; a hypothesis that we plan to pursue in future studies.

B11B-0740 0830h POSTER

Subsurface Microbial Communities and Geochemistry Within a Vertical Transect of a Uranium-Contaminated Aquifer

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Microbial communities and geochemistry were analyzed within floodplain alluvia of the San Juan River, New Mexico, down-gradient of a uranium mill tailings disposal cell. A multi-level sampling device was used to investigate vertical variations in geochemistry and microbial community structure within the uranium contaminant plume within this shallow aquifer.

Chemical analyses indicate that the interaction of uncontaminated artesian water and contaminant fluids has resulted in three geochemically-distinct regions. Similarity comparisons of bacterial community 16S rDNA fingerprints, based on T-RFLP analyses, show a grouping of microbial populations into three assemblages. These groupings correspond to the three geochemically-defined regions of the aquifer profile, indicating a relationship between community structure and geochemistry. Combined Bacterial 16S rDNA clone library and T-RFLP analyses show a predominance of organisms related to Nitrospira and Nitrosolobus, chemolithotrophic nitrite and ammonia oxidizers respectively, in the uncontaminated region of the aquifer profile. Within the plume-impacted area, organisms related to known nitrifying bacteria were not detected. Bacteria phylogenetically related to Acinetobacter, Aeromonas, Gallionella, Pseudomonas, and Thiomicrospira were identified in both the uncontaminated and plume regions. Within the contaminated region, sequences from organisms related to the metal oxidizing bacteria Leptothrix and Sphaerotilus were also found. 16S rDNA sequences with high similarity to Duganella zoogloeoides, a chemolithotrophic Mn-oxidizing bacterium known for uranyl complexation and sorption, were abundant in the clone library from the plume-impacted region. A diversity of organisms related to sulfate- and sulfur-reducing bacteria including Desulfobulbus, Desulfofrigus, Desulfosarcina, Desulfosporosinus, Desulfotomaculum, and Geobacter were present in the uncontaminated zone while less SRB diversity (Desulfobacter, Desulfobulbus, and Desulfonema) was observed in the plume region. T-RFLP results substantiate this trend, suggesting sulfate reducer populations shift from high diversity and low relative abundance to less diversity and a larger abundance over the transition from uncontaminated water into the plume region. These findings indicate that there is considerable biological potential for uranium immobilization at this site through reductive precipitation by sulfate or other metal reducing organisms or microbial uptake and sequestration. Given the diversity of microorganisms related to chemolithotrophic bacteria, growth by this mode of metabolism, and the biological oxidation of reduced metals, is also potentially an important process that may influence metal biogeochemistry at this site.

B11B-0741 0830h POSTER

Microbial Diversity of Groundwater from Deep Subsurface Environment

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The subsurface environment harbors one of the most abundant reservoirs of biomass on Earth. The distribution of microbial ecosystems and the diversity of microbial metabolisms there remained poorly understood due to lack of detailed sampling over three-dimensional space with extremely heterogeneous characteristics. South African Au mines, however, provide the best access in the world to various types of groundwater and rocks at depths up to 4 km below surface. In this study, we present our recent analyses of microbial community structure of groundwater (with residence time of several million years) collected from depths between 850 to 1500 mbsl of Beatrix Au mine, South Africa.

Five groundwater samples were collected anaerobically from freshly drilling boreholes with flow rates of 1 to 38 L/min. Cells were concentrated through filtration and total DNA were extracted from filters and PCR-amplified with primers targeting 16S rDNA gene. The amplicons were cloned and digested with restriction enzymes to identify the unique clone type. Sequences were obtained through direct sequencing of representative clones and compared with the closest matching sequences deposited in the gene bank for the construction of phylogenetic tree.

The archaeal signatures were only found in one sample and close to the lineage of methanosarcina. The most predominant ribotype was similar to the environmental clone found in the same mine under the species level while the rest of ribotypes were either close to those capable of methanogenesis from long-chain alkanes or found in rice field or were distant from other environmental clones reported in previous study (Takai et al., 2001). The bacterial community exhibited a wide range of diversity among samples. Most samples were dominated by sequences close to alpha proteobacteria with various proportions of beta, gamma proteobacteria and environmental clones. A significant proportion of sequences close to thermophilic delta proteobacteria and clostridia were observed from one of the deepest samples. Since the in-situ water or rock temperature at sampling location is below the temperature range for thermophilic bacteria, this might indicate that the microorganisms once colonizing in the deeper and hotter portion of crust were transported upward with hydrothermal fluid and preserved in the sealed water pocket for a time scale of millions of years.

B11B-0742 0830h POSTER

In-situ Microbial Cultivation in a South African Gold Mine

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An in-situ culturing device, with four individual enrichment cartridges, was developed to be installed and incubated anaerobically for one month, in a flowing borehole at a depth of 1.81 km in a South African gold mine. The design of the experiment was based on preliminary microbial phylogenetic characterization and culturing results obtained from a South African gold mine water sample. The device was developed in order to harvest methanogenic, iron-reducing, and sulfate-reducing microorganisms, typically resistant to cultivation, within their natural environment thus increasing their biomass enough so as not to be overtaken by more dominating microorganisms. An additional goal of the device was to establish a correlation between the phylogenetic data and the actual physiology of the microbial constituents. Preliminary geochemical results indicate that the device quickly became anaerobic as flow was forced through the enrichment cartridges due to a pressure release/flow regulating valve system. Upon retrieval of the cartridges, the agar and sand portions were set aside for lipid and DNA extractions, cation and anion analyses, sulfur isotope analysis, and inoculations at 32C, the in-situ temperature. In addition, at the same time that the cartridges were retrieved, water samples were collected for dissolved gas and other geochemical analyses. Initial inoculation results indicate positive Ferredoxin and methanogenic cultures. We anticipate this device to prove useful in future attempts to examine unculturable microbes and to help correlate molecular phylogenetic data with microbial physiology.

B11B-0743 0830h POSTER

Hydrocarbon Gas and Microbial Geochemistry in the Deep Subsurface of the Witwatersrand Basin, South Africa

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Field investigations in the deep mines of South Africa, targeting gas and fluids under extreme conditions of temperature, pressure, salinity, and locally radioactivity indicate the existence of micro-organisms (in particular methanogens). Gases associated with the saline water pockets (>3km in depth) within the Precambrian Witwatersrand Basin, South Africa are rich in hydrocarbons (methane, ethane, propane and butane). Compositional analyses and stable carbon and hydrogen isotopes of individual hydrocarbon components are used to identify the origin of the gases. Based on the depleted $\delta^{13}\text{C}$ signatures and high methane/high hydrocarbon ratios, 38-100% of the gas encountered in the Witwatersrand quartzite and shale sequences is estimated to be microbial in origin. In contrast, an isotopic pattern consistent with abiogenic gas is found in samples from the Ventersdorp lava sequences, although some degree of mixing with microbial gases is also indicated. Mixing between abiogenic and microbially produced hydrocarbons explains the range of isotopic signatures and compositions obtained for the samples. Confirmation of the presence of methanogens based on microbial evidence will be presented.

B11B-0744 0830h POSTER

Carbon isotopes of lipid biomarkers in modern travertine deposits: Implications for biogeochemical dynamics in hot-spring systems (Yellowstone National Park, USA)

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The goal of this study was to understand carbon cycling pathways in the context of changing depositional environments in modern travertine deposits. Microbial mats were collected for lipid and carbon isotope studies along a temperature gradient in four depositional facies at Angel Terrace, Mammoth Hot Springs, Yellowstone National Park. Bacterial phospholipid fatty acids (PLFA) appeared to be responsive to temperature variation. The vent facies (71°C) was dominated by unsaturated 20:1 (32%) and 18:1 (17%) fatty acids and saturated 18:0 (22%) fatty acid, which reflect bacterial adoption to high temperature. The down-gradient facies (the pond, 51°C; the proximal slope, 47°C; and the distal slope, 24°C) were dominated by saturated fatty acid 16:0 (30-55%), which exceeds that in the vent (10%). On the other hand, 20:1 was less than 1% in these relatively low-temperature environments. Total organic carbon (TOC) and PLFA in vent facies were significantly enriched in C-13 compared to those in the down-gradient facies. For example, the carbon isotope ratio in the vent was -13.7 per mil for TOC and -18.7 per mil for 16:0. In the pond and proximal slope, these values were about -27 and -36 per mil, respectively. The distal slope saw a slight increase in C-13 with TOC of -23.5 per mil and 16:0 of -28.9 per mil. The more positive C-13 values are consistent with isotope fractionation pathways performed by thermophilic sulfide-oxidizing Aquificales, which are primary producers in the vent. The more negative C-13 values in the pond, proximal slope and distal slope are typical of cyanobacteria and other autotrophic bacteria predominant in these low-temperature settings. These results may provide insight into biogeochemical dynamics in ancient travertine deposits with varying depositional facies.

B11B-0745 0830h POSTER

Dead or Alive? Probing Microbial Ecosystems With Intact Polar Lipids.

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Multi-proxy approaches consisting of isotopic information derived from diagnostic lipid biomarkers together with phylogenetic techniques have advanced our understanding of naturally occurring microbial communities. Examples include the significant advances in our understanding of the ecology of archaeal methanotrophs and planktonic crenarchaeota. However, this approach has to be modified for application to microbial ecosystems in the deep subsurface in order to distinguish live matter from dead. Polar lipids, i.e., phospho- and glycolipids, fulfill that requirement and potentially provide information on the structure, size and function of the microbial community. We utilized a high-performance-liquid-chromatograph coupled to a

mass-spectrometer (HPLC-MS) to examine the distribution of intact polar lipids from microbial isolates and surface sediments that are relevant for the study of deep subsurface biospheres. Structural features of bacterial and archaeal-derived polar lipids are identified on the basis of their sequential fragmentation in an ion-trap mass spectrometer upon electrospray ionization. Additional details are obtained from a series of chemical degradations designed to release alkyl units of the polar lipids rendering them analyzable by conventional gas-chromatography-mass-spectrometry (GC-MS). Fingerprints of polar lipids from microbial isolates allow distinction of taxonomically closely related prokaryotes. Moreover, sediments from seep environments in Eel River Basin and Guaymas Basin that host anaerobic methanotrophic communities are both dominated by similar types of glycosyl biphityanyl tetraethers suggestive of a predominant low-diversity archaeal assemblage. First applications of this technique to sediments from the deep subsurface will be presented.

B11B-0746 0830h POSTER

Interaction of *Pseudomonas fluorescens* with Eu(III) and Ce(IV) Desferrioxamine Complexes

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Naturally occurring chelating agents, such as siderophores, are able to form complexes with actinides and enhance their solubility and mobility in the environment. Adsorption and/or biodegradation of chelated actinides by microorganisms are important processes which regulate their mobility in the natural environment.

In this study, association of Eu(III), Ce(IV), and Fe(III) desferrioxamine B (DFO) complexes with aerobic bacterium, *Pseudomonas fluorescens* (ATCC 55241), was investigated. Eu(III) and Ce(IV) were used as analogues to trivalent and tetravalent actinides, respectively. When 20 μ M of 1:1 Eu(III) - and Ce(IV) DFO complexes were incubated with *P. fluorescens* in 0.1 M Tris-HCl buffer (pH = 7.3), the metals were removed from solution, with no change in DFO in solution. With decreasing metal/DFO molar ratio from 1 to 0.01, the accumulation of Eu(III) and Ce(IV) by *P. fluorescens* decreased. Kinetics study showed that accumulation of Eu(III) reached the maximum within 30 minutes, and then it decreased slightly with time. On the other hand, Ce(IV) accumulation proceeded in a parabolic process where the kinetics was slower than that of Eu(III) accumulation. In comparison to Eu(III) and Ce(IV), the removal of Fe(III) added as a DFO complex by *P. fluorescens* was not observed. The formation constants (log K) of Eu(III) - DFO and Fe(III) - DFO are reported to be 15 and 30.6, respectively.

These results suggest that Eu(III) DFO complex was dissociated in the presence of bacteria cells and was readily biosorbed.

B11B-0747 0830h POSTER

ATR-FTIR Spectroscopic and Ab-initio Theoretical Studies of the Coordination Geometry of Oxalate Anions at Mineral-Water Interfaces.

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Oxalate is the smallest dicarboxylate ligand and one of the most abundant low-molecular weight organic acids in nature, originating from secretions by plant roots, bacteria and fungi in rhizospheres of plant roots. In addition to its structural simplicity and natural abundance, oxalate also has a strong adsorption affinity for mineral surfaces and is known to catalyze mineral dissolution reactions. We have studied the coordination geometry of oxalate anions adsorbed at boehmite (g-AlOOH)/water and corundum (a-Al₂O₃)/water interfaces using in-situ ATR-FTIR spectroscopy combined with ab-initio quantum chemical geometry optimizations and IR vibration frequency calculations.

To attain a more realistic simulation of the oxalate-aluminum oxide complex, we used an AlO₁₂ cluster as the adsorption substrate and conducted calculations using HF/6-31+G*(3-21G) basis sets for three possible surface complexation models, including (a) bidentate side-on coordination with 5-membered ring formation, (b) monodentate end-on coordination without ring formation, and (c) bidentate end-on coordination with 4-membered ring formation. Model (a) is energetically preferred to models (b) and (c). It is also preferred to a simplified model of the hydrated aluminum oxide surface, in which one water molecule is bonded to a surface Al atom. Further IR frequency calculations using models (a), (b), and (c) and comparison with the observed data support the above energetic considerations; the predicted frequencies for model (a) match very well with the FTIR spectra of the oxalate-boehmite and oxalate-corundum samples, while those from models (b) and (c) show significant deviations. From this comparison of theoretical experimental results, we conclude that the most probable coordination geometry for individual oxalate anions adsorbed on aluminum-(oxyhydr)oxides is bidentate side-on coordination with 5-membered ring formation. This finding helps explain the strong observed uptake of oxalate on aluminum-(oxyhydr)oxides and its role in the dissolution of metal oxides.

B11B-0748 0830h POSTER

Mechanisms of Eu(III) and Cm(III) Association With *Chlorella Vulgaris*

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Association of Eu(III) and Cm(III) with *Chlorella vulgaris* and cellulose was studied by a batch method, time-resolved laser-induced fluorescence spectroscopy (TRLFS) and extended X-ray absorption fine structure (EXAFS). The kinetics study performed by a batch method showed that the maximum adsorption of Eu(III) and Cm(III) on *C. vulgaris* was attained within three minutes of contact time, and afterwards the percentage adsorption decreased with time due to exudates released from *C. vulgaris* with affinity for Eu(III) and Cm(III). TRLFS showed that the short-term adsorption of Eu(III) on *C. vulgaris* was attributed to their coordination with the cell wall components comprised of cellulose. TRLFS also demonstrated that Eu(III) coordinated with the functional groups of cellulose very weakly in spite of the large distribution coefficients observed. EXAFS analysis showed the local structure around the Eu(III) adsorbed on cellulose and with *C. vulgaris* was similar. These results indicate that the reactions both at cell surfaces through the adsorption as well as in solution phases through chelation with the exudates are important in estimating the environmental behavior of Eu(III) and Cm(III) in aqueous environments.

B11B-0749 0830h POSTER

Cytochrome c Biogenesis Genes Involved in Arsenate Respiration by *Shewanella trabarsenatis* ANA-3

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Arsenate can be used as a terminal electron acceptor in anaerobic respiration by diverse bacteria. The detection of these bacteria in numerous contaminated environments suggests that they are widespread and metabolically active in nature. Arsenate-respiring bacteria have been implicated in the mobilization of arsenic from arsenic-contaminated sediments. However, the enzymatic mechanisms supporting arsenate respiration are largely unknown. Here, we describe c-type cytochromes that are involved in arsenate respiration by the bacterium *Shewanella trabarsenatis* strain ANA-3, a facultative anaerobe that is able to use a variety of electron acceptors for growth. We performed transposon mutagenesis to study the electron transport pathway in ANA-3 during arsenate respiration. 10 arsenate-respiration deficient mutants were found after screening up to 7,000 mutants, and 4 were shown to have unique

transposon insertions through Southern Blot analysis. The physiological properties of these mutants were determined, including characterization of their growth on different electron acceptors. The genes flanking the transposon insertions were sequenced for each mutant, and several were found to encode c-type cytochrome biogenesis genes. UV/VIS spectra and SDS/PAGE were used to confirm the absence of c-type cytochromes in the mutants. Based on these findings, we proposed a model for respiratory electron transport to arsenate.

B11C MCC: Hall C Monday 0830h Water, Energy, and Carbon Exchange in Forest Systems III Posters (joint with A, H, GC)

Presiding: B Law, Oregon State University; P Thornton, National Center for Atmospheric Research; D Baldocchi, University of California, Berkeley

B11C-0750 0830h POSTER

Spatial Variability of Carbon Fluxes and Soil Respiration Within the Understory of a Pacific Northwest Old-Growth Forest

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We present data and analysis on the spatial heterogeneity of carbon fluxes measured by 2 additional eddy covariance systems in the forest understory at the Wind River Canopy Crane AMERIFLUX site. The Crane is located in a Pacific Northwest Old-growth Forest with trees up to 500 years old and 65 meters tall. A permanent eddy covariance station consisting of a Gill-Solent HS Research sonic anemometer and a LiCor LI6262 closed-path InfraRed Gas Analyzer (IRGA) has been operated for over 4 years at a height of 2.5 meters to complement an identical system measuring total ecosystem exchange at a height of 70 meters. Two additional eddy covariance systems, consisting of Campbell CSAT-3 sonic anemometers and open-path IRGAs (LiCor 7500 and NOAA probe) were located in the understory along a 200 m transect for a period of 13 months. The forest structure is complex with seven gymnosperm and two angiosperm tree species in the 2.3 ha crane circle, large amounts of woody debris on the forest floor, and a diverse understory. Soil respiration is a major contributor to the carbon budget at the site. The long-term understory Eddy-covariance data indicate the release of carbon from the soil to be as large as 11 tC ha⁻¹ yr⁻¹ with maximum values of 6 to 8 μ mol m⁻¹ s⁻¹. The Net ecosystem carbon exchange (NEE) estimated by eddy-covariance ranges from a sink of 1.9 tC ha⁻¹ yr⁻¹ to a source of 0.5 tC ha⁻¹ yr⁻¹. In this study we investigate the spatial and temporal dynamics of carbon fluxes within the canopy understory.

B11C-0751 0830h POSTER

of Horizontal Advection of Carbon Dioxide Within a Forest Canopy

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We present methodology, data and analysis on the horizontal advection of Carbon Dioxide within the understory at the Wind River Canopy Crane AMERIFLUX site. The Crane is located in a Pacific Northwest Old-growth Forest with trees up to 500 years old and 65 meters tall. The forest structure is complex with seven gymnosperm and two angiosperm tree species in the 2.3 ha crane circle, large amounts of woody debris on the forest floor, and a diverse understory. Data presented was collected using a 3-dimensional CO₂/H₂O profile system using LiCor LI6262 and LI7000 closed-path InfraRed Gas Analyzers (IRGA) with a total of