

most prevalent form of land-use change in the region over the study period has been the transformation of Pithaya forest and coastal wetlands to shrimp aquaculture. Shrimp farms that did not exist in the region in the early 1980s now represent over 12 percent of the study area. Our analysis suggests that this boom in shrimp farming was influenced by a series of policy reforms instituted by the Mexican Government over the last decade intended to open the rural economy to global markets. These reforms include modifications to the Fisheries, Foreign Investment and Land Tenure Laws, changes in the rural credit system, and liberalization of international trade policies (NAFTA). The data indicate overall increased rates of land conversion from natural covers (Pithaya forest, Mesquite forest, Choyal, salt flats) to human dominated ecosystems (aquaculture, agriculture, salt ponds, urban) in the post-reform period (1994 - 2001) compared to the pre-reform period (1973 - 1992). Our results highlight the importance of monitoring local impacts in evaluating national policies.

#### B21A-05 0930h

##### Carbon Dynamics in the Southeastern Plains: A Preceding Study to Quantify the Consequences of Contemporary Land Cover and Land Use Change on Terrestrial Carbon Stocks and Fluxes in Conterminous U.S.

Shuguang Liu<sup>1</sup> ((605)594-6168; sliu@usgs.gov)

Thomas R Loveland<sup>2</sup> ((605)594-6066; loveland@usgs.gov)

Rachel M Clement<sup>2</sup> ((605)594-6118; rclement@usgs.gov)

<sup>1</sup>U.S. Geological Survey, EROS Data Center, Raytheon, Sioux Falls, SD 57198, United States

<sup>2</sup>U.S. Geological Survey, EROS Data Center, Sioux Falls, SD 57198, United States

Coupled with the USGS/EPA study on the spatial and temporal dimensions of contemporary U.S. land cover and land use change (LCLUC), we assess how changes in land cover and land use affect local, regional and national carbon stocks and fluxes in terrestrial ecosystems. A low cost sampling strategy based on Omernik ecoregions is used to localize estimates of the rates of LCLUC and to quantify the corresponding consequences on carbon dynamics. Sampling blocks were randomly selected for each of the 84 ecoregions to identify > 1% change in cover within each ecoregion at an 85% confidence level. The analysis of land cover change is based on five dates of Landsat MSS, TM, and ETM data (nominally 1973, 1980, 1986, 1992, and 2000). Using an ensemble approach, we deploy the CENTURY ecosystem model to simulate carbon dynamics within each of the sampling blocks at the resolution of 60 m by 60 m. The goal is to identify the spatial distribution and temporal change of carbon sources and sinks in the conterminous U.S. and explain the mechanisms that cause the variability. In this paper, our carbon simulation approach is presented with the southeastern plains as a case study.

Part of this work is performed under U.S. Geological Survey contract 1434-CR-97-CN-40274.

#### B21A-06 0945h

##### Land Use Effects on Net Greenhouse Gas Fluxes in the US Great Plains: Historical Trends and Model Projections

Stephen J Del Grosso<sup>1</sup> (970 491 1919;

delgro@nrel.colostate.edu); William J Parton<sup>1</sup> (970 491 1988; billp@nrel.colostate.edu); Dennis S Ojima<sup>1</sup> (970 491 1976; dennis@nrel.colostate.edu); Arvin R Mosier<sup>1,2</sup> (970 490 8250; amosier@lamar.colostate.edu); Keith Paustian<sup>1</sup> (970 491 1547; keithp@nrel.colostate.edu); Gary A Peterson<sup>3</sup> (970 491 6804; gpeterso@lamar.colostate.edu)

<sup>1</sup>Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, CO 80523-1499, United States

<sup>2</sup>USDA/ARS, P.O. Box E, Fort Collins, CO 80522, United States

<sup>3</sup>Soil and Crop Science, Colorado State University, Fort Collins, CO 80523-1170, United States

We present maps showing regional patterns of land use change and soil C levels in the US Great Plains during the 20<sup>th</sup> century and time series of net greenhouse gas fluxes associated with different land uses. Net greenhouse gas fluxes were calculated by accounting for soil CO<sub>2</sub> fluxes, the CO<sub>2</sub> equivalents of N<sub>2</sub>O emissions

and CH<sub>4</sub> uptake, and the CO<sub>2</sub> costs of N fertilizer production. Both historical and modern agriculture in this region have been net sources of greenhouse gases. The primary reason for this, prior to 1950, is that agriculture mined soil C and resulted in net CO<sub>2</sub> emissions. When chemical N fertilizer became widely used in the 1950's agricultural soils began to sequester CO<sub>2</sub>-C but these soils were still net greenhouse gas sources if the effects of increased N<sub>2</sub>O emissions and decreased CH<sub>4</sub> uptake are included. The sensitivity of net greenhouse gas fluxes to conventional and alternative land uses was explored using the DAYCENT ecosystem model. Model projections suggest that conversion to no-till, reduction of the fallow period, and use of nitrification inhibitors can significantly decrease net greenhouse gas emissions in dryland and irrigated systems, while maintaining or increasing crop yields.

#### B21A-07 1000h

##### Observational Constraints From Local to Continental Scale on the Analysis of an Integrated Regional Carbon Model

Dennis S. Ojima (970-491-1976; dennis@nrel.colostate.edu)

Colorado State University, Natural Resource Ecology Laboratory Colorado State University, Fort Collins, CO 80523-1499, United States

During the past decade, terrestrial ecologists have been in the search of "the missing carbon sink" which atmospheric scientists have suggested resided in the ecosystems of the northern hemisphere. This search has led to refinements in our basic understanding of carbon cycling and how terrestrial ecosystems modify the changing concentrations of atmospheric CO<sub>2</sub>. The quantification of the changes in carbon fluxes from different terrestrial ecosystem due to climate change and human activities such as deforestation, fire suppression, and agricultural practices at local to regional scales is critical to our understanding of how we are affecting greenhouse gas emissions and how we may reduce the rate of these emissions. Regional analysis of biological control on the terrestrial biospheric processes controlling carbon exchange with the atmosphere and storage of carbon within the ecosystem will greatly enhance our ability to quantify carbon and other greenhouse gas emission from regions within the United States of America and globally. The integrated analysis of land, atmosphere, and human dimension processes provide insight to the factors contributing to the changes in atmospheric CO<sub>2</sub> and the biospheric role in affecting source sink changes on seasonal to inter-annual timeframes. Multiple observations of fluxes, concentrations, and states of C provide constraints on the integrated regional C analysis. Inventory analyses and simulation results indicate that land use has a major impact on the net carbon biome sink of the conterminous US in the range of 0.2 Gt per year. Development of an integrated regional C model includes the processes related to ecosystem development, such as regrowth patterns of forests, plant composition changes, impact of fire, grazing, and cultivation on different ecosystems.

#### B21B MC: 120 Tuesday 0830h

##### Assessing Bioremediation I (joint with H)

Presiding: J P McKinley, Pacific

Northwest National Laboratory; F S Colwell, Idaho National Engineering and Environmental Laboratory

#### B21B-01 0830h INVITED

##### Principles of Bioremediation Assessment

Eugene L Madsen (607 255 3086; elm3@cornell.edu)

Department of Microbiology, Cornell University, Ithaca, NY 14853, United States

Although microorganisms have successfully and spontaneously maintained the biosphere since its inception, industrialized societies now produce undesirable chemical compounds at rates that outpace naturally occurring microbial detoxification processes. This presentation provides an overview of both the complexities of contaminated sites and methodological limitations in environmental microbiology that impede the documentation of biodegradation processes in the field.

An essential step toward attaining reliable bioremediation technologies is the development of criteria which prove that microorganisms in contaminated field sites are truly active in metabolizing contaminants of interest. These criteria, which rely upon genetic, biochemical, physiological, and ecological principles and apply to both in situ and ex situ bioremediation strategies include: (i) internal conservative tracers; (ii) added conservative tracers; (iii) added radioac-

tive tracers; (iv) added isotopic tracers; (v) stable isotopic fractionation patterns; (vi) detection of intermediary metabolites; (vii) replicated field plots; (viii) microbial metabolic adaptation; (ix) molecular biological indicators; (x) gradients of coreactants and/or products; (xi) in situ rates of respiration; (xii) mass balances of contaminants, coreactants, and products; and (xiii) computer modeling that incorporates transport and reactive stoichiometries of electron donors and acceptors. The ideal goal is achieving a quantitative understanding of the geochemistry, hydrogeology, and physiology of complex real-world systems.

#### B21B-02 0845h INVITED

##### Effects of Subsurface Microbial Ecology on Geochemical Evolution of a Crude-Oil Contaminated Aquifer

Barbara A Bekins<sup>1</sup> (650-329-4691; babekins@usgs.gov)

Isabelle M. Cozzarelli<sup>2</sup> (703-648-5899; icozzare@usgs.gov)

E. Michael Godsy<sup>1</sup> (650-329-4504; emgodsy@usgs.gov)

Ean Warren<sup>1</sup> (650-329-4554; ewarren@usgs.gov)

Frances D. Hostettler<sup>1</sup> (650-329-4584; fdhostet@usgs.gov)

<sup>1</sup>U.S. Geological Survey (USGS), MS 496 345 Middlefield Rd., Menlo Park, CA 94025, United States

<sup>2</sup>USGS, 431 National Center, Reston, VA 20192, United States

We have identified several subsurface habitats for microorganisms in a crude oil contaminated located near Bemidji, Minnesota. These aquifer habitats include: 1) the unsaturated zone contaminated by hydrocarbon vapors, 2) the zones containing separate-phase crude oil, and 3) the aqueous-phase contaminant plume. The surficial glacial outwash aquifer was contaminated when a crude oil pipeline burst in 1979. We analyzed sediment samples from the contaminated aquifer for the most probable numbers of aerobes, iron reducers, fermenters, and three types of methanogens. The microbial data were then related to gas, water, and oil chemistry, sediment extractable iron, and permeability. The microbial populations in the various contaminated subsurface habitats each have special characteristics and thus affect the aquifer and contaminant chemistry.

In the eight-meter-thick, vapor-contaminated vadose zone, a substantial aerobic population has developed that is supported by hydrocarbon vapors and methane. Microbial numbers peak in locations where access to both hydrocarbons and nutrients infiltrating from the surface is maximized. The activity of this population prevents hydrocarbon vapors from reaching the land surface. In the zone where separate-phase crude oil is present, a consortium of methanogens and fermenters dominates the populations both above and below the water table. Moreover, gas concentration data indicate that methane production has been active in the oily zone since at least 1986. Analyses of the extracted separate-phase oil show that substantial degradation of C<sub>15</sub>-C<sub>35</sub> n-alkanes has occurred since 1983, raising the possibility that significant degradation of C<sub>15</sub> and higher n-alkanes has occurred under methanogenic conditions. However, lab and field data suggest that toxic inhibition by crude oil results in fewer acetate-utilizing methanogens within and adjacent to the separate-phase oil. Data from this and other sites indicate that toxic inhibition of acetoclastic methanogenesis in the proximity of separate phase contaminant sources may result in build-up of acetate in contaminant plumes.

Within the aqueous-phase contaminant plume steep vertical hydrocarbon concentration gradients are associated with sharp transitions in the dominant microbial population. In the 20 years since the aquifer became contaminated, sediment iron oxides have been depleted and the dominant physiological type has changed in areas of high contaminant flux from iron reducing to methanogenic. Thus, methanogens are found in high permeability horizons down gradient from the oil while iron reducers persist in low permeability zones. Expansion of the methanogenic zone over time has resulted in a concomitant increase in the aquifer volume contaminated with the highest concentrations of benzene and ethylbenzene.

URL: <http://www.mn.cr.usgs.gov/bemidji/>

#### B21B-03 0900h INVITED

##### In-situ evidence for uranium immobilization and remobilization

Lee R Krumholz<sup>1</sup> (405-325-0437; krumholz@ou.edu)

John M Senko<sup>1</sup> (405-325-4011; senko@ou.edu)

Jonathan D Istok<sup>2</sup> (541-737-8547; Jack.Istok@orst.edu)

Joseph M Sufilita<sup>1</sup> (405-325-5761; jsufilita@ou.edu)

<sup>1</sup>University of Oklahoma, Department of Botany and Microbiology and Institute for Energy and the Environment, Norman, OK 73019

<sup>2</sup>Oregon State University, Dept. of Civil Engineering, Corvallis, OR 97331

The in-situ microbial reduction and immobilization of uranium was assessed as a means of preventing the migration of this element in the terrestrial subsurface. Uranium reduction and microbial respiratory activities were evaluated in the presence of exogenous electron donors and acceptors with field push-pull tests using wells installed in an anoxic aquifer contaminated with landfill leachate. Uranium(VI) amended at 1.5  $\mu$ M was reduced to less than 1 nM in groundwater in under 8 d during all field experiments. Amendments of 0.5 mM sulfate or 5 mM nitrate slowed U(VI) immobilization and allowed for the recovery of 10% and 54% of the injected element, respectively compared to 4% in the unamended treatment. Laboratory incubations confirmed the field tests and showed that the majority of the U(VI) immobilized was due to microbial reduction. In these tests, nitrate treatment (7.5 mM) not only inhibited U(VI) reduction, but also appeared to remobilize uranium as nitrite was transiently produced. This latter finding was confirmed with field push-pull tests in which 0.6 mM or 7.5 mM nitrate and 1.5 mM U(VI) were injected into sediments which already contained immobilized uranium. After an initial loss of the amendments, the concentration of soluble U(VI) increased and eventually exceeded the injected concentration, indicating that previously immobilized uranium was remobilized as nitrate was reduced. Laboratory experiments using heat-inactivated sediment slurries suggested that the denitrification intermediates, nitrite, nitrous oxide, and nitric oxide, were all capable of oxidizing and mobilizing U(IV). These findings indicate that in-situ subsurface U(VI) immobilization can be expected to take place under anaerobic conditions, but the permanence of the approach can be impaired by denitrification intermediates that can mobilize previously reduced uranium.

**B21B-04 0915h INVITED**

**Assessing Enhanced Anaerobic and Intrinsic Aerobic Biodegradation of Trichloroethene**

Kent S. Sorenson<sup>1</sup> ((208) 528-8718 x129; ksorenson@nwindenv.com)

Roger L. Ely<sup>2</sup>

Jennifer P. Martin<sup>3</sup>

Lisa Alvarez-Cohen<sup>4</sup>

Mary E. Kauffman<sup>5</sup>

<sup>1</sup>North Wind Environmental, Inc., P.O. Box 51174, Idaho Falls, ID 83405, United States

<sup>2</sup>Yale University, Dept. of Chemical Engineering, New Haven, CT, United States

<sup>3</sup>Idaho National Engineering and Environmental Laboratory, Geosciences Research, Idaho Falls, ID, United States

<sup>4</sup>University of California - Berkeley, Dept. of Civil and Environmental Engineering, Berkeley, CA, United States

<sup>5</sup>Idaho National Engineering and Environmental Laboratory, Biotechnology, Idaho Falls, ID, United States

Biodegradation of chloroethenes can proceed either anaerobically or aerobically; however, the techniques for monitoring the two pathways are quite different. At the Idaho National Engineering and Environmental Laboratory's Test Area North (TAN), a combination of anaerobic and aerobic biodegradation of trichloroethene (TCE) is being employed for restoration of a large plume of contaminated groundwater. During stimulation of anaerobic biodegradation of TCE through lactate addition, several assessment tools have proven effective for various objectives. Monitoring TCE and its lesser chlorinated degradation products provides a straightforward assessment tool for the occurrence of degradation. It does not, however, provide information regarding the potential for reductive dechlorination, nor progress from less suitable to more suitable conditions. A technique for obtaining this information is monitoring redox-sensitive geochemical parameters such as dissolved iron, sulfate, methane, and oxidation-reduction potential. This approach was demonstrated by the strong correlation of steps in the reductive dechlorination pathway to redox conditions at the TAN site. Yet another tool is required to determine adequacy of conditions for efficient dechlorination. Dechlorination efficiency appears to be dependent upon the predominant electron donor utilization (or fermentation) process occurring at any given time, an observation consistent with thermodynamic considerations. Thus, monitoring of added electron donor and intermediate product concentrations can help determine an efficient operations strategy. One final tool demonstrated at the TAN site was monitoring stable carbon isotope ratios. As TCE was dechlorinated, a clear fractionation occurred from cis-dichloroethene to

vinyl chloride, and from vinyl chloride to ethene. This fractionation provides a clear signature of reductive dechlorination.

Assessment of aerobic biodegradation of chloroethenes at TAN is more challenging because of the lack of readily identifiable degradation products. The aerobic pathway for TCE consists of cometabolic oxidation by any of several oxygenase enzymes. Intermediates generated in this process are very short-lived. Thus, monitoring of degradation products in the field has not been found to be useful. While disappearance of TCE might suggest aerobic biodegradation, it does not distinguish biodegradation from other processes such as dilution. This issue was resolved at TAN through normalizing TCE concentrations to the concentrations of internal plume tracers. This allowed degradation to be distinguished from physical processes such as dispersion, dilution, sorption, and volatilization. When combined with standard techniques such as enumeration and screening of organisms in site groundwater with the capability to aerobically degrade TCE, this technique provided convincing evidence that intrinsic aerobic biodegradation is occurring at the site, and estimated a rate. Direct measurement of this process remains a challenge, but preliminary work with activity-dependent, fluorescent enzyme probes appears promising. While the appropriate assessment tool depends on the objectives to be met, a combination of tools will almost always be necessary for thorough documentation and implementation of biodegradation of chloroethenes in the field.

**B21B-05 0930h**

**Use of Biotracer Tests to Evaluate the Feasibility of In-Situ Bioremediation**

Fara J Lourenco<sup>1</sup> (flourens@hwr.arizona.edu)

Mark L Brusseau<sup>1</sup> (brusseau@ag.arizona.edu)

<sup>1</sup>The University of Arizona, Shantz Bldg. #38, Room 429 PO BOX 210038, Tucson, AZ 85721, United States

In-situ bioremediation of organic compounds in the subsurface is increasing in popularity. A biodegradation tracer test, also known as biotracer test, is one approach to determine the feasibility of practicing this method. This test involves conducting a tracer experiment with one or more compounds whose biodegradation properties are known. The mass recovery and transport of the biotracers are compared to a non-biodegradable, conservative tracer to measure the biological activity of the target zone. Field experiments were conducted at three sites. All three sites were contaminated with non-aqueous phase liquids. Biotracers were also used to investigate the response of the system to the addition of perturbations, e.g. the addition of oxygen. The results from these field experiments suggest biotracers are a promising method for characterizing in-situ bioremediation.

**B21B-06 0945h**

**In Situ Respiration and Direct Enzymatic Assays for Assessing Bioremediation**

Terry C. Hazen (510-486-6223; tchazen@lbl.gov)

Lawrence Berkeley National Laboratory, MS 70A-3317 One Cyclotron Rd., Berkeley, CA 94720, United States

Laboratory and field studies of in situ bioremediation at sites contaminated with organics or metals have demonstrated that in situ measurements of respiration using start/stop tests or He tracers combined with direct assays of dehydrogenase activity in the sediment are much more effective in determining the efficacy of the bioremediation strategy being used. Sites with high TOC (> 6000 ppm) can have a low oxygen demand if the carbon present is not bioavailable or is recalcitrant to the indigenous microorganisms, as indicated by both respiration rate and dehydrogenase activity. Addition of surfactants or limiting nutrients at these sites increases both the respiration rate and the dehydrogenase activity resulting in a concomitant decrease in the contaminants of concern (TPH, PAH, TCE, DCE, VC, MSW). Similar studies in the laboratory have shown that reduction of chromium is correlated with increased dehydrogenase activity and not to densities of microorganisms or the presence of particular groups as indicated by molecular probes or fatty acid analyses. In situ respiration tests and direct enzymatic measurements of sediment may provide better control of in situ bioremediation processes by indicating need for addition of nutrients, surfactants, or additional carbon/energy supplements.

**B21C MC: 120 Tuesday 1020h**

**Biological Mineralization: Proxies and Processes (joint with OS, PP, MR)**

**Presiding:** P M Dove, Virginia

Polytechnic Institute and State

University; J J DeYoreo, Lawrence

Livermore National Laboratory

**B21C-01 1020h INVITED**

**Mineralization of the Sea Urchin Skeleton**

Fred Wilt (510-642-2807; wilt@socrates.berkeley.edu)

University of California, Berkeley, Molecular Cell Biology Department, 142 LSA, Berkeley, CA 94720-3200, United States

The sea urchin possess a calcareous skeleton composed of over 99% magnesian calcite, an enveloping extracellular matrix, and an occluded protein matrix. The most intensively studied skeletal element is the spicule of the embryo. At the 32 cell stage of development a cohort of 4 cells becomes irrevocably dedicated to spicule formation. At the early gastrula stage the descendants of these founder cells form the primary mesenchyme (PMC). The PMCs fuse to form a multi-nucleated syncytium connected by cytoplasmic cables, and the calcitic skeleton is formed within these cables. Our primary concern is with the cellular and molecular mechanisms that support the formation of the mineralized spicules. The import of calcium into the PMCs results in appearance of intracellular vesicles containing precipitated calcium, which is neither very stable nor birefringent, and could be amorphous. The precipitated calcium is vectorially secreted into an extracellular space. This space is almost completely enclosed by cytoplasmic strands, and the mineral is encased in an extracellular matrix. Proteins destined for the extracellular matrix, and for inclusion in the spicule, are present in the Golgi membranes and in small intracellular vesicles. These vesicles apparently deliver the matrix proteins to the growing spicule. Our current view is that the matrix molecules are much more than a passive armature, but are actively involved in precipitation, secretion, and organization of the mineral phase.

**B21C-02 1035h INVITED**

**Silica Synthesis by Sponges: Unanticipated Molecular Mechanism**

Daniel E. Morse<sup>1</sup> ((805) 893-8982; d\_morse@lifesci.ucsb.edu)

James C. Weaver<sup>1</sup> ((805) 893-3416; weaver@lifesci.ucsb.edu)

<sup>1</sup>University of California, Santa Barbara, Marine Biotechnology Center, Marine Science Institute, Santa Barbara, CA 93106, United States

Oceanic diatoms, sponges and other organisms synthesize gigatons per year of silica from silicic acid, ultimately obtained from the weathering of rock. This biogenic silica exhibits a remarkable diversity of structures, many of which reveal a precision of nanoarchitectural control that exceeds the capabilities of human engineering. In contrast to the conditions of anthropogenic and industrial manufacture, the biological synthesis of silica occurs under mild physiological conditions of low temperatures and pressures and near-neutral pH. In addition to the differentiation between biological and abiotic processes governing silica formation, the biomolecular mechanisms controlling synthesis of these materials may offer insights for the development of new, environmentally benign routes for synthesis of nanostructurally controlled silicas and high-performance polysiloxane composites.

We found that the needle-like silica spicules made by the marine sponge, *Tethya aurantia*, each contain an occluded axial filament of protein composed predominantly of repeating assemblies of three similar subunits we named "silicateins." To our surprise, analysis of the purified protein subunits and the cloned silicatein DNAs revealed that the silicateins are highly homologous to a family of hydrolytic enzymes. As predicted from this finding, we discovered that the silicatein filaments are more than simple, passive templates; they actively catalyze and spatially direct polycondensation to form silica, (as well as the phenyl- and methyl-silsesquioxane) from the corresponding silicon alkoxides at neutral pH and low temperature. Catalytic activity also is exhibited by the silicatein subunits obtained by disaggregation of the protein filaments and those produced from recombinant DNA templates cloned in bacteria. This catalytic activity accelerates the rate-limiting hydrolysis of the silicon alkoxide precursors. Genetic engineering, used to produce variants of the silicatein molecule with substitutions of specific amino acid sidechains, in conjunction with computer-assisted