Appendix A

Agenda

Workshop on Research Opportunities in Biogeochemical Dynamics

U.S. Department of Energy
Natural & Accelerated Bioremediation Research Program
August 4-5, 1998

Doubletree Hotel at the Quay
100 Columbia Street
Vancouver, Washington

Background

Program Goal

The primary goal of the NABIR Program is to develop a fundamental scientific basis for intrinsic and accelerated *in situ* bioremediation of metals and radionuclides at DOE sites. A key to the success of the program will be understanding the biologically mediated geochemical processes of importance in controlling contaminant behavior over the broad range of geohydrologic conditions at DOE sites. Major challenges include defining the principal biogeochemical processes operating in the subsurface at DOE sites, implementing a program to understand the controls on the rate and direction of these processes, and extrapolating this information to the field.

Biogeochemical Dynamics Workshops

Previous workshops were undertaken to survey bioremediation field technologies and to evaluate the manipulation of biogeochemical processes as a strategy for extending the range of subsurface conditions available for scale-up of laboratory research to the field. This workshop will build on the results of prior workshops and current NABIR research and begin the process of further defining and prioritizing research needed to understand the biogeochemical processes that influence the behavior of metals and radionuclides in heterogeneous subsurface environments.

Workshop Objectives

Refine themes for focusing research on key biogeochemical processes underlying bioremediation of metals and radionuclides and explore collaborative opportunities for future research to understand how these processes are manifested at the field scale.

Research Themes

The term “research theme” refers to a scientific topical area of extraordinary benefit to field-scale bioremediation, in which there is (1) strong potential for scientific progress (major advances in the state-of-the-art are needed), and (2) collaborative, cross-disciplinary research promises substantial scientific advancements compared to individual investigator studies. Research themes have bioremediation focal points (e.g., reductive immobilization of metals) and address two or more subsidiary research questions that must be answered in order to understand how the process is manifested in heterogeneous subsurface environments.

Proposed research themes include:

- Microbial transformation of subsurface mineral phases and effects on contaminant mobilization/immobilization (dissolution/desorption or promotion of strongly sorbed, occluded, or coprecipitated contaminants)
- Microbially mediated complexation processes (effects of complex formation/degradation on contaminant aqueous/solid phase interactions)

Subsidiary questions center on the effects of heterogeneity on the distribution, rate, and direction of these processes (mass transfer controls on microbial distribution and nutrient and electron donor/acceptor dynamics). The workshop will consider the state of the art in research on these processes and experimental approaches to understand the effects of heterogeneity and manipulation on their rate and direction in the field.

Workshop Approach

- Review the results of previous workshops to provide perspective on the range of environments, primary research issues, and possible use of core-scale approaches for extrapolating mechanistic studies to the field.
- Summarize latest NABIR research on the research themes (principal investigators from Biogeochemical Dynamics Program Element) and experimental resources/approaches for examining process coupling and upscaling (principal investigators from other NABIR program elements and visitors).
- Based on the above, assess the relevance of the themes, scientific needs, and potential integrated experimental approaches for translating process-level understanding to natural systems.
- Explore opportunities for collaboration among investigators to address technical gaps.

August 4

7:30 - 8:00 am **Continental Breakfast**

8:00 - 8:30 am **Session I - Introduction (Scribe: D. White)**

The objective of this session will be to provide the background, goals, and proposed approach for the workshop.

- NABIR Biogeochemical Dynamics Program Element Needs. F. Wobber
- Workshop Goals and Desired Products (candidate themes/collaborative approaches to guide research within limited resources) F. Wobber

8:30 -10:00 am **Session II - Perspective**
(Moderator: R. Wildung; Scribes: E. Roden/S. Brooks)

The objective of this session is to review the results of previous NABIR workshops to provide insights into scientific issues and possible approaches for describing biogeochemical processes at the field scale. The session will describe the range in operating and environmental conditions at DOE field sites that influence bioremediation strategies, general research issues in heterogeneity, and the opportunities and limitations of biochemical manipulation and core scale approaches to address these issues that evolved from previous workshops sponsored by the Biogeochemical Dynamics Program Element. Workshop summaries will be distributed in advance of the meeting.

- Workshop Themes and Approach. R. Wildung

- Gaps in Field Knowledge/Technologies for Bioremediation Research (report on the workshop Field Technologies for Bioremediation Research). P. Long

- Manipulation/Core Scale Approaches for Upscaling Biogeochemical Processes (report on the workshop Biogeochemical Manipulation for Scale-Up of Laboratory Research to the Field). J. Zachara

10:00 - 12:00 pm **Session III - Current Knowledge of Biogeochemical Processes** (Moderator: R. Crawford; Scribes: J. Wall, T. Onstott)

The objective of this session is to summarize the results of NABIR research on the principal biogeochemical processes operating in the subsurface. The session will provide investigators with the opportunities to share the latest results and react individually to perspectives offered in Session II for relating the results of their research to the field.

Presentations (20 min) will be by investigators in the Biogeochemical Dynamics Program Element and include brief updates on objectives and key results and investigator views on collaborative opportunities to address heterogeneity/field extrapolation issues identified in Session II.

- Reductive Solubilization (synthetic iron oxides, contaminated sediments). J. Zachara/ J. Fredrickson

- Direct and Metabolite Induced Reduction (iron oxides). S. Fendorf

- Sulfate Reduction (sediments, intact cores). J. Suflita

- Dissimilatory Reduction of Technetium. R. Wildung

- Nitrate Effects on Oxidation-Reduction (iron oxides, sediments). F. Picardal/E. Roden

- Biodegradation of Metal Complexes and Scale-up (structured saprolites). S. Brooks/P. Jardine

12:00 - 1:00 pm **Lunch**

1:00 - 2:00 pm **Session III - Biogeochemical Processes (Continued)**

2:00 - 5:30 pm **Session IV - Perspectives from Other Program Elements and Investigators** (Moderator: D. White; Scribes: J. Suflita, P. Jardine)

The objective of this session will be to gain the insights of investigators from other NABIR program elements and visiting scientists, focusing on subsurface heterogeneity and resources/ approaches relevant to extrapolating microbially mediated processes to the field.

Presentations (20 min) will emphasize interdisciplinary opportunities for collaborative research.

Core/Microcosm Scales

- Microbial Resources for Investigation of Biogeochemical Processes Representing Different Hydrogeologic Regimes - SMCC East and West. D. Balkwill/D. Boone (see footnote)

- Microbial Distribution in Relation to Physical and Geochemical Properties. F. Brockman

Intermediate Scale

- Intermediate-Scale Approaches for Scaling Microbial Processes in Flow Regimes. E. Murphy

Field Scale

- Determining Heterogeneity in Physical Properties at the Field Scale. E. Majer

- Innovative Imaging and Tracer Technology for Scaling Biogeochemical Processes. G. Moline

- Single and Dual Well Injection/Withdrawal Approaches for Measuring Microbial Dynamics in the Field. R. Crawford

- Iron Reduction and Bacterial Transport in Unconsolidated Sandy Sediments: Scaling Concepts. T. Onstott/T. Scheibe

5:30 - 5:45 pm	**Assignments for next session, if any** (R. Wildung, R. Crawford, D. White)

August 5

7:30 - 8:00 am	**Continental Breakfast**
8:00 - 12:00 pm	**Session V - Collaborative Research to Address Technical Gaps** (Moderators: J. Zachara , G. Moline; Scribes R. Wildung, R. Crawford, D. White)

The objective of this session will be to achieve a general closure on experimental themes, approaches, and collaborative opportunities for extending biogeochemical process-level understanding to the field. A roundtable discussion will include the scribes from Sessions II, III, and, IV who will briefly summarize the conclusions from discussions in these sessions as they relate to the research themes and then participate in a discussion to refine research needs and collaborative approaches to address those needs.

Emphasis will be on the following questions:

- Do the experimental themes (and subsidiary questions related to heterogeneity) address the most critical biogeochemical processes for intrinsic and accelerated bioremediation? Are there other themes/processes operational over a range of DOE sites with greater potential for fundamental scientific progress and benefits to bioremediation?

- Based on the current state-of-the-art in biogeochemistry, what are the pivotal issues that must be addressed in extending mechanistic studies at the microscopic, grain, and pore scales in the laboratory to larger scales in the field where heterogeneity may play a dominant role? For example, is it possible to estimate:

  - slow rate processes often typical of field environments?

  - effects of fluid fluxes on nutrient and electron donor acceptor availability and removal of metabolic products?

  - the role of heterogeneity in these processes?

- What approaches or combination of approaches, such as those described in Session II or IV, offer the highest probability for success in addressing key issues?

- What advances in measurement, experimental, and modeling approaches are needed to scale laboratory-based research to the field?

- What are the opportunities for collaboration within and outside the NABIR program?

- What additional capabilities/skills are needed to fill scientific gaps and implement research?

12:00 - 1:00 pm **Lunch**

1:00 - 3:30 pm **Session V (Continued)**

3:30 - 4:30 pm **Session VI - The Path Forward**

- Perspectives on Collaborative Research Needs (Session V). R. Crawford, D. White, R. Wildung

- Proposed Actions. F. Wobber

Adjourn

Footnote: The anaerobic culture collection (SMCC -West) is located at Portland State University. A visit to view the collection and research facilities can be arranged on Monday, August 4.

Appendix B

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Appendix C

Summary of Workshop Discussion

This appendix summarizes the scribe notes for discussions in each of the major workshop sessions (Appendix A). Notes are generally organized according to the questions on collaborative research to address research needs posed to participants in Session V.

Session II. Perspective

Research Themes

- Experimental themes proposed were generally deemed to be scientifically appropriate. However, during the discussion, a suggestion was raised that the concept of scaling could itself become a research theme—into which various biogeochemical processes involved in contaminant remediation would be enfolded.

- The outcome of a scaling-driven theme could provide invaluable information for designing bioremediation processes over a range of DOE sites.

- With the concept of scaling firmly in place, it would be desirable to work toward development of a "universal bioremediation strategy," whose basic framework could be applied to many different types of DOE sites.

- The idea of a "universal bioremediation strategy" evolved from the description of the integrative thinking that went into development of the hypothetical "universal contaminated site" on DOE lands.

Pivotal Issues

- The issues of *slow field reaction rates and fluid flux* can be dealt with in a tractable way in experimental systems; first step in this direction is use of intact cores.

- Regarding the issue of *heterogeneity* (which is of course a huge issue): the "flip side" of using intact cores to explore influence of micro-mesoscopic heterogeneity on biogeochemical processes is that adequate methodologies for characterizing that heterogeneity will be required.

- There is a need to characterize heterogeneity in intact experimental cores (proposed at the January 1998 intact core workshop) using intact cores for "archeological" assessment of how heterogeneity may be related to the fate of contaminant metals/radionuclides in subsurface materials. In essence, if the heterogeneity of a core segment can be analyzed, then this information can be correlated with information on the association of contaminants with different features within that heterogeneity in order to understand how heterogeneity affects the fate of contaminants.

- While the "archeological" approach makes sense conceptually, it was cautioned that it is possible that heterogeneities, e.g., in contaminant distribution, which one might try to explain based on observed physical/chemical heterogeneities within a given core segment could actually have been caused by processes working on a much large scale.

- Concerns about the issue of *intact core disturbance*, as well as *difficulties in obtaining adequate replicates and controls*, are relevant to the above discussion. Important to note here, though, that these issues do not necessarily represent "show-stoppers" in terms of using intact cores for biogeochemical research. Must bear in mind, however, that there is apparently no reliable "benchmark" upon which to base assessments of the impact of this issue.

- Regarding the issue of techniques for dealing with *scaling* questions as they relate to the issue of heterogeneity, the presentations indicated that considerable progress has been made (e.g., through application of "streamtube theory") in dealing with the influence of physical heterogeneity on hydrological transport and its attendant impact on soluble organic contaminant biodegradation.

- Major challenge at present is to learn to deal with heterogeneity associated with solid-phase chemical and microbiological properties. Some theoretical progress is being made in this direction.

Approaches

- Investigators seemed to agree that the intact core approach is a compelling alternative and has a high probability of success; however, there are many caveats and potentially very important limitations that must be considered along the way.

- In spite of the potential limitations, there seems to be a strong consensus that intact cores will likely provide the best opportunity for obtaining specific, detailed information on the role of (1) chemical/microbiological heterogeneity, and (2) unique effects of coupled transport (including bacteria!) and reaction, on biogeochemical processes related to metal/radionuclide transformation.

- Other clear consensus regarding intact core research: it should in all cases be hypothesis-driven and scientifically well-grounded! The basis for this conclusion is established in the report of the workshop in January 1998.

- The considerable success NABIR researchers have had using intact cores and soil blocks will likely provide a wealth of advice and expertise to others wishing to pursue this approach.

- Intermediate-scale flow cells allow control of fluid flow and have the advantage that predetermined levels of physical/chemical heterogeneity can be built in, allowing for experimental assessment of the influence of such heterogeneity on biogeochemical processes. Clearly, this approach is more time consuming and expensive, and thus the questions that can be addressed will be more limited in scope than may be achievable with intact cores.

- Field scale is, of course, the most relevant to real-world remediation; ultimately all roads must lead in this direction.

Advances Needed

- Obvious major need is need to be able to characterize heterogeneity at various spatial scales; perhaps the two most important scales discussed during this and ensuing sessions were the level of intact cores and at the field scale.

- The most challenging need for advancement in the realm of experimental approaches is development of effective techniques for working with intact cores; as mentioned earlier, issues of disturbance, slow rates, and replication are substantial hurdles, but hopefully not show-stoppers.

- The single most important thing to be done at this point may be for individual investigators to begin to use and contribute to development of intact core methodology.

- With regard to modeling, at least two major issues can be identified: (1) dealing with problems of heterogeneity and scaling to the field, and (2) availability of user-friendly kinetic/equilibrium models for use in mechanistic biogeochemical research.

- The former issue was dealt with specifically in several presentations and investigators seem to be making clear progress.

- The latter issue apparently remains a problem for many biogeochemical investigators, particularly the more microbiologically oriented ones who are not that familiar with how to work with powerful but non-user-friendly routines. A lengthy discussion was held on this general topic. One major conclusion that emerged was that it is not likely that having a general-purpose biogeochemical modeler on the NABIR biogeochemical team is a good approach; in other words, bringing in someone to help people with their modeling needs would likely not be a successful strategy. Rather, modeling collaborations should be established up-front and take place in integrated manner with experimental work.

Opportunities for Collaboration

- The suggestion of 2-3 relatively large, integrated research programs directed toward field-scale research makes sense, as does the suggestion that the programs be led by a hydrogeologically oriented investigator—i.e., someone who can take into account the various scales at which heterogeneity and its influence on biogeochemical processes may manifest in the field.

- There is greater need for collaboration between experimental mechanistic researchers and modelers.

Additional Needs

- Two items in addition to those already emphasized above emerged at various times:

  - importance of characterizing microbiological heterogeneity in relation to physical/chemical heterogeneity must be explicitly emphasized.

  - potential for reversibility of biogeochemical-based bioremediation strategies needs to be considered; for example, the possibility for reoxidation of metal-sulfide precipitates, leading to remobilization of contaminant metals.

Session III. Current Knowledge of Biogeochemical Processes

(Refer also to Appendix D for concise summaries of presentations)

Research Themes

- A proposed theme that encompasses pertinent issues in biogeochemistry might be: What is the impact of physical and chemical heterogeneity on microbially mediated complexation and mineralization?

- Proof of concept for difficult experiments. Both SRB and DIRB activity can immobilize metals (in oxides and sulfides) in batch and stirred reactor experiments. Soluble electron acceptors like nitrate may inhibit DIRB activity or lead to precipitation of Fe oxides. The use of XAS *in situ* analyses of aqueous speciation promises to unravel details of sorption and complexation mechanisms at the pore scale and the short-term stability of complexes. These types of experiments could be carried forward ad infinausea with different strains, and consortia, different media and minerals, and are probably performed by many other investigators outside NABIR and the U.S.

- But how well do these processes transfer to porous or fractured media in natural groundwater systems? Is this where biogeochemical dynamics wants to establish leadership?

Pivotal Issues

- What is the long-term geological survival of the precipitated oxide or sulfide phases?
- How do we assess activity *in situ*, i.e., how do we know if it is working or if it is not working, why it is not working?
- Do protists play a role?
- Do we investigate the effect of consortia versus pure strains?
- Role of humic acids or other electron shuttles?
- How important are microgradients in pH at the mineral/bacteria/fluid interface and how do we assess that?

Approaches

- *In-situ* activity or biomass measurements tested in intact core or in intermediate flow cells for potential use down the hole? For example, is there an approach for *in-situ* activity measurements for DIRB activity?
- Rates—does the addition of humic analogs, which enhance rates in lab, provide an estimate of total reducibility over the long term? Keeping in mind that you also have to scale for biomass concentration.
- Fluid fluxes—this requires downhole measurements of flow velocities and colloid transport velocities and concentration gradients.
- Heterogeneity—spatial heterogeneity in intact core seems feasible. Down the hole processes for determining heterogeneity like static MLS's, *in-situ* microcosms, or push-pull interrogation give spatial resolution, but limited temporal resolution. What temporal resolution do we need at the field scale?
- Modelling—can we benefit from evolutionary models for biotransformation of metals or the role of consortia?

Opportunities for Collaboration

- New technologies are too expensive to develop—need off the shelf. New technologies are out there searching for an application, e.g., Lincoln Laboratory down-hole CPT UV laser-down-the-hole, high-resolution soluble organics.

Additional Needs

- Hot spots (heterogeneity): What are the causes? Is it a physical configuration and limitation? Chemical? Biological? Is there an underlying heterogeneity that would predict a hot spot? Are there a combination of factors at work? Are there specific associations of microbes at active sites of bioreduction that are important?
- Are there other metals that should be considered that are now not being looked at? Should more complex mixtures be investigated at this stage?
- Only geothite is receiving much attention as a solid substrate. Should other minerals, silicates, or clays be studied?
- What is the effect of grazing on the microbiological community that is expected to mediate the biogeochemistry? What is the protist community in these environments?
- Humics are suggested to play a role in mediating extracellular electron transfer, but what is the distribution of humics in the environment in which biogeochemical reactions need to take place? Quinone amendments tested?
- Stability of reduced species? Abiotic stability? Biotic stability?

- Studies of FRB and SRB are reported. Are other bacteria important? Fermentors? Acetogens? Methanogens? Denitrifiers?
- Major need for a visualization technique for FRB like the silver foil technique for SRBs.
- Almost all experiments with growing bacteria. That is not the situation *in situ*. What are the effects of stress on the processes under study?

Session IV. Collaborative Research to Address Technical Gaps

(Refer also to Appendix E for concise summaries of presentations)

Research Themes

Research themes encompass primary biogeochemical processes influencing metal/radionuclide behavior in subsurface environments.

Pivotal Issues

- How does physical heterogeneity (e.g., high and low permeability distributions) impact the transport and distribution of microbes and contaminants?
- How does geochemical heterogeneity (e.g., mineralogical distribution) impact the transport and distribution of microbes and contaminants?
- How does water connectivity and degree of saturation affect the distribution and survivability of microbes and the kinetics of microbially mediated processes?
- How do rate-limiting processes such as diffusion, sorption, and redox reactions impact the bioavailability of contaminants and nutrients?
- Can core-scale observations of coupled geochemical, hydrological, and microbial processes be used to resolve field-scale issues involving biogeochemical dynamics?

Approaches

- Measurement of geologic, hydrologic, geochemical, and microbial characteristics from the field site where undisturbed cores are extracted.
- Preliminary core characterization using nondestructive interrogation techniques to image the internal structure and composition of the core.
- Application of various molecular, biochemical, and microbiological techniques to elucidate the structure of microbial communities.
- Application of manipulative multitracer experiments for distinguishing geochemical and physical heterogeneities as well as time-dependent mass transfer processes vs. equilibrium-based reactions.
- Application of manipulative experiments to demonstrate the effectiveness of microbial degradation or reductive processes under a range of different environmental conditions.
- Application of manipulative experiments for demonstrating the effectiveness of biostimulation techniques for enhancing microbial contaminant transformation reactions.
- Application of numerical modeling techniques that consider the statistical variability of coupled geochemical, hydrological, and microbial processes involved in contaminant biotransformation and remediation.

Advances Needed

- Noninvasive measurement technologies on cores

  - CT scans (media structure and distribution)

  - Small-angle-neutron-scattering (SANS - media structure and scaling)

  - Fiber optics (chemical and microbial distributions)

  - Synchrotron Sources (XAS, contaminant redox, and binding mechanisms/ distributions, real-time measurements)

  - *In situ* microprobe techniques (pH, DO, redox couples)

- Experimental technologies on cores

  - Multiple flow rates and pressure-heads to alter flow path dynamics

  - Multiple tracers with different diffusion coefficients and sizes to quantify coupled time-dependent processes

  - Linking the above with fiber optics, time-resolved synchrotron sources, and microprobes.

- Modeling

  - Fractal approaches using SANS and CT coupled with macroscopic transport observations

  - Stochastic approaches using SANS, CT, XAS coupled with macroscopic transport observations.

Collaborative Opportunities

- Collaborative Oyster/field research needs (available resources identified in Appendix E)

  - Geophysical characterization

  - Multitracer strategies/ hydrogeochemistry

  - Geostatistical analysis of data

  - Application of existing scaling theory

- Consider common use of microorganisms available in the SMCC.

- Advanced flow and tracer technologies are available for an extended core-scale research effort.

- Intermediate-scale concepts/approaches and facilities are available to address questions of scaling in conjunction with core and field studies.

Appendix D

Current Knowledge of Subsurface Biogeochemical Processes

The objective of Session III was to summarize NABIR biogeochemical research on the principal biogeochemical processes controlling metal/radionuclide behavior in the subsurface. Brief synopses of the presentations on projects currently in the Biogeochemical Dynamics Program Element are given below:

Iron Reduction

- Reductive Solubilization (synthetic iron oxides, contaminated sediments). J. Zachara/J. Fredrickson

  Central Hypothesis: Advective removal of Fe(II) will stimulate dissimilatory Fe reduction

  Approach: Investigations in batch and stirred flow reactors are being undertaken to examine microbial (*Shewanella*, Geothrix) reductive solubilization of contaminants associated with Fe oxides and the effects of advective removal of Fe(II) on the stability of immobilized metals.

  Key Initial Results: Natural Fe oxides from sediments are more bioreducible than synthetic oxides.

- Dissimilatory Reduction of Technetium. R. Wildung

  Central Hypothesis: Technetium mobility in anaerobic nonsulfidogenic environments is controlled by iron reduction and complexation of reduced forms with inorganic and organic ligands of natural and waste origin.

  Approach: Direct Tc and indirect [biogenic Fe(II)] reduction mechanisms and kinetics are being evaluated in batch culture and the effects on Tc speciation (XAS), equilibrium distribution with the solid phase and mobility in anaerobic columns are being determined and modeled.

  Key Initial Results: Tc reduction and speciation is a function of electron donor type and solution composition. Soluble and insoluble hydrolytic species were identified that control Tc solubility. Tc reduction in the presence of carbonate led to the formation of highly electronegative dimeric Tc species. Soluble and insoluble species were shown to be consistent with thermodynamic predictions.

- Nitrate Effects on Oxidation-Reduction (iron oxides, sediments). F. Picardal/E. Roden

  Central Hypothesis: Iron reduction and the subsequent effects on metal behavior will be influenced by competitive interactions between nitrate and Fe(III) oxide reduction and nitrate-dependent Fe(II) oxidation.

  Approach: Microbial consortia from natural waters are being used in batch equilibrations to test the effect of nitrate on Fe reduction and reoxidation.

  Key Initial Results: Nitrate (5 mM) inhibits microbial reduction of goethite and the incorporation of Zn into the solid phase. Nitrate oxidizes both aqueous and solid phase Fe(II).

Iron/Sulfate Reduction

- Direct and Metabolite Induced Reduction (iron oxides). S. Fendorf

Central Hypothesis: Iron and sulfur-reducing organisms will directly reduce contaminants if Fe(III) and sulfate are not bioavailable. Chemical reduction will occur when sufficient Fe and S are available.

Approach: Chemical and biological mechanisms (*Shewanella, Desulfovibrio*) of reduction of Co(III)EDTA, Cr(VI), and U(VI) are being examined in stirred, anaerobic batch reactors containing solid, reduced Fe(II) and S(II) phases. Analyses are by XAS (which may be directly linked to the reaction vessels) and FTIR.

Key Initial Results: Uranyl reduction by *Shewanella* was greater in the absence of Fe(III) and with goethite as the source of Fe(III) compared to ferrihydrite, which contained more bioavailable Fe. Sulfide will reduce Co(III)EDTA to Co(II(EDTA). $Fe(OH)_3$ will reduce Cr(VI) through a mixed Fe-Cr amorphous product, releasing Fe(II), which is subsequently oxidized by Cr(III).

Sulfate Reduction

- Sulfate Reduction (sediments, intact cores). J. Suflita/L. Krumholz

  Central Hypothesis: Sulfate reduction will result in immobilization of metals/radionuclides.

  Approach: Sulfate reduction (*Desulfovibrio, Desulfoarcula, Desulfomicrobium*) and metal behavior is addressed in batch and intact cores, including direct autoradiographic imaging of precipitated sulfide. Log normal distribution of sulfate in low sulfate reducing cores and normal distribution in high sulfate reducing cores suggesting localized activity in sediment niches.

  Key Initial Results: Co(II) is precipitated as sulfide and is dependent on sulfate concentration and electron donor.

Complexation Scale-Up

- Biodegradation of Metal Complexes and Scale-up (structured saprolites). S. Brooks/P. Jardine

  Central Hypothesis: Biodegradation of metal chelating agents will be diffusion-limited and a function of pore size.

  Approach: Laboratory and field observations of chelate degradation and metal transport in porous media.

  Key Initial Results: Stability of organic (NTA) complexes of Co to degradation are a function of Co oxidation state and reoxidation processes; Co(III) complexes are more stable to degradation than Co(II) complexes and oxidation of Co(II) in NTA occurs in the presence of saprolites.

Appendix E

Upscaling Biogeochemical Processes in Heterogeneous Systems: Resources and Approaches

The objective of Session IV was to gain the insights of investigators from other NABIR program elements and visiting scientists, focusing on subsurface heterogeneity and resources/approaches relevant to extrapolating microbially mediated processes to the field. Presentations emphasized interdisciplinary opportunities for collaborative research and are summarized below.

Core (mm to dm) Scale

- Microbial Resources for Investigation of Biogeochemical Processes Representing Different Hydrogeologic Regimes—SMCC East and West. D. Balkwill/D. Boone

  - Investigators from throughout the NABIR program are taking advantage of the collection and services in microbial screening and distribution, characterization and preservation organisms and training in anaerobic techniques. Access to organisms from different sites facilitates the extrapolation of the results of biogeochemical studies to the range of environments typical of the DOE complex.

  - New opportunities exist at the new Oyster field site to augment strains in the collection and to focus on organisms (e.g., iron reducers) that are of unique potential importance in governing metal/radionuclide behavior.

  - As in previous field studies on the Subsurface Science Program, the physical and chemical properties of the environments from which the organisms are being isolated at the Oyster site have been highly characterized. This provides a basis for interpretation of the presence and abundance of microorganisms in the context of the field environment from which they were isolated, including the factors controlling distribution and the dominant biogeochemical processes occurring under different subsurface conditions.

- Innovative Imaging and Tracer Technology for Scaling Biogeochemical Processes. G. Moline

  - New insights into pore-scale dynamics, process interactions and controls in intact cores may be obtained using intact cores.

  - Approaches offering real potential include a combination of multiple tracer techniques to define sorption and matrix diffusion and x-ray computed tomography for noninvasive imaging of fracture aperture, spacing, and length scales.

Intermediate (dm to m) Scale

- Intermediate-Scale Approaches for Scaling Microbial Processes in Flow Regimes. E. Murphy

  - Intermediate-scale flow cells provide an experimental approach for scaling biogeochemical processes to the field. Operating at the meter scale, experiments of this type can capture the effects of non-uniform convection that dominate spreading in a plume though control and quantification of the interactive effects of physical and geochemical heterogeneity and fluid flow on microbially mediated processes and microbial transport.

- Numerical strategies (deterministic, averaging, and stochastic) have been developed to address a range of biogeochemical scaling issues, from situations in which averaging parameters over the field scale is a legitimate approach (e.g., adsorption/desorption reactions that vary linearly with concentration) to situations in which averaging parameters may fail (e.g., as may occur in biologically mediated processes where reaction rate may be nonlinear and dependent on other parameters).

Field (dm) Scale

■ Microbial Distribution in Relation to Physical and Geochemical Properties. F. Brockman

- The distribution of microbes is markedly influenced by physical and geochemical properties in subsurface sediments and geostatistical approaches offer strong insights into the spatial dependence of key parameters that influence subsurface microbial processes.

- Intensive vertical and horizontal sampling at the centimeter to meter scale has revealed the critical nature of sample size and how averaging scale changes with hydraulic connectivity, sediment age and history, and nutrient limitations.

■ Determining Heterogeneity in Physical Properties at the Field Scale. E. Majer

- A number of noninvasive and invasive geophysical techniques (surface, borehole logging, surface to borehole, borehole to borehole) are now available for interrogating subsurface media, including static and dynamic properties and scaling from point to volume. The method required will depend on the objective, scale of heterogeneity, and the required level of resolution.

- The field studies at Oyster exemplify how geophysical imaging can be linked with manipulative experiments to better understand subsurface processes.

■ Single and Dual Well Injection/Withdrawal Approaches for Measuring Microbial Dynamics in the Field. R. Crawford

- The push-pull technique offers a tool for characterizing and microbial communities in real time and under actual groundwater conditions.

- The experiment involves several phases, including purging and sampling of wells, injection of test solutions, drift in which the test materials move with the groundwater flow, and withdrawal in which returned solute and reaction products are quantified.

- The basic approach can be utilized in a number of ways to provide insights into the dynamics of subsurface processes. Examples include (1) coupling both reactive and nonreactive tracers to provide insights into the important physical and geochemical processes that may control microbial abundance and distribution, (2) nutrient or electron donor injection to examine impacts on microbial communities or contaminant removal/transformation without impact on subsurface systems downgradient, and (3) use of dual wells and dual microbial injections to examine interactions between microbial communities.

■ Iron Reduction and Bacterial Transport in Unconsolidated Sandy Sediments: Scaling Concepts. T. Onstott/T. Scheibe

- Scaling concepts and approaches have been developed to represent observations at one scale in a corresponding effective process at a larger scale, addressing the effects of heterogeneity in properties at intervening scales. Focus has been on bacterial transport in a heterogeneous sandy aquifer at the Oyster site.

- Scaling approaches have included (1) simplified conceptual models that assume perfect stratification, lumped equilibrium partitioning processes, and no parameter transfer across scales; (2) peturbation methods that assume an idealized model of spacial heterogeneity, small variance and correlation between physical and chemical properties; (3) Monte-Carlo Stochastic Simulation in which high-resolution property fields are generated from geostatistical or geologic process models and measurements at appropriate scales; and (4) stochastic-convective-reactive methods (utilized in intermediate-scale studies, above) in which flow fields are represented by an ensemble of streamtubes, and hydrology is described through inert tracer behavior.

- The studies at the Oyster field site are directed toward examining the role of Fe-Al-Mn oxyhydroxides, hypoxia, and Fe reduction on bacterial retention and far-field transport.

- Preliminary laboratory studies with intact (50 cm) cores and materials from the Oyster site suggest that sediment effects will be important in influencing bacterial transport and predictive models may have to take into account the effects of small (mm) scale physical and geochemical (hydrous Fe/Al oxide) heterogeneities on bacterial retention.