Integrated Field, Laboratory, and Modeling Studies to Determine the Effects of Linked Microbial and Physical Spatial Heterogeneity on Engineered Vadose Zone Bioremediation

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Linkages to DOE Site Needs

Remediation of chlorinated hydrocarbons in deep vadose zones of arid climates

- 340,000 - 450,000 liters CCl_4 resides in Hanford vadose zone
- Majority of contaminant mass is in the vadose zone at Hanford

In situ bioremediation can offer advantages in cost, speed, public acceptance, & final cleanup level achieved, but ...

Uncertainties exist in applying in situ bioremediation to DOE vadose zone problems

- How effectively can nutrients be delivered to sparse & spatially heterogeneous microbial populations in deep vadose zones?
- Predictive capabilities are immature for engineered in situ bioremediation of chlorinated hydrocarbons in deep vadose zones.
- How much site characterization information is needed for parameterizing reactive transport models to enable accurate predictions of vadose zone processes?

Objective

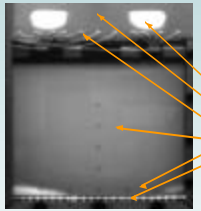
Address key knowledge gaps that currently limit our ability to assess the feasibility of engineered bioremediation of chlorinated compounds in *heterogeneous, microbially sparse vadose zones*.

Specific Objectives:

1) Laboratory evaluation of approaches for biotransformation of CT
   - Gas-phase nutrient injection (no water or organism additions)
   - Point injections of nutrient solutes and organisms
2) Develop an improved vadose zone reactive transport model
   - Laboratory research focused on understanding interacting hydrologic & microbiological processes
   - Incorporation of microbial processes into STOMP model
3) Develop guidelines on the level of site characterization needed for accurate modeling of engineered vadose zone bioremediation
   - Detailed characterization of properties at field site
   - Reactive transport simulations with varying degrees of site information as input

2-D Vadose Visualization Chamber

45 x 50 x 1 cm volume



- Reference windows for light bank
- UV lamp housing
- Influent drip emitters
- Injection and sampling ports
- Effluent manifold

How Much Site Characterization is Needed for Accurate Modeling of Bioremediation

Data source: Extensive characterization of uncontaminated vadose zone site at Hanford

Modeling approach: Characterization data applied to a *hypothetical saturated zone* TCE/phenol plume undergoing intrinsic bioremediation

Case 1 -- Average biomass from field data applied homogenously throughout simulation domain

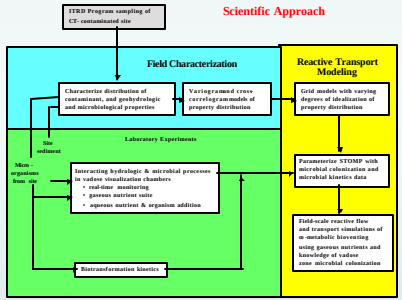
Case 2 -- Heterogeneous biomass distribution applied to simulation domain by random sampling of histogram of field data

Case 3 -- Heterogeneous biomass distribution applied to simulation domain by using full set of correlated permeability-biomass data from field.

Modeling Assumptions

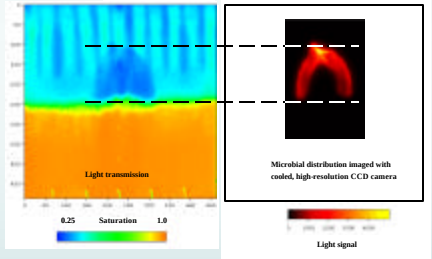
- Advective-dispersive transport with kinetic reaction sources/sinks (RAFT code)
- Biomass (attached to aquifer solids - no transport)
  - 0.1% of initail biomass capable of TCE co-metabolism
  - Triple-Monod growth rate term; first-order death rate term
- Phenol (primary substrate; only aerobic degradation)
  - Triple-Monod utilization rate term for biomass growth
  - Dual-Monod utilization rate term for metabolism
- Dissolved oxygen (electron acceptor)
  - Stoichiometrically related to phenol utilization rate
- Phosphate as limiting nutrient
  - Cycling included both readily-available & eventually-available forms
  - Cycling based on cellular phosphate & cell growth/death rates
- TCE degradation (aerobic co-metabolism)
  - Degraded at rate proportional to phenol utilization
  - Competitively inhibited at higher phenol concentrations

Scientific Approach

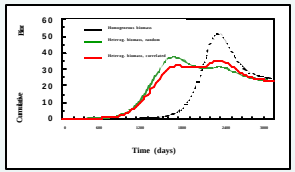


Project research activities (large colored boxes), research tasks (small boxes), and flow of materials and information (arrows)

Direct, noninvasive observation of microbial distribution and activity (right) with simultaneous monitoring of water content distributions and flow paths (left) in unsaturated porous media (images from Oregon State Univ. NSF project)

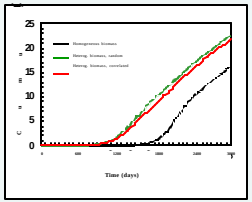


Simulated Biomass History



- Biomass accumulation was delayed in homogeneous case
- Random sampling of histogram was sufficient

Simulated TCE Degradation

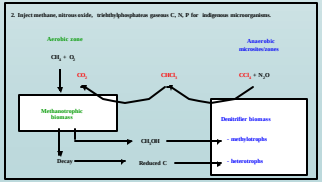


- TCE degradation was delayed and less in homogeneous case
- Random sampling of histogram was sufficient

Approaches for CT Transformation

1. Inject nutrients to create anaerobic microzones/zones; inject *Pseudomonas stutzeri* strain KC to convert CCl_4 to CO_2 without buildup of chloroform ($CHCl_3$).

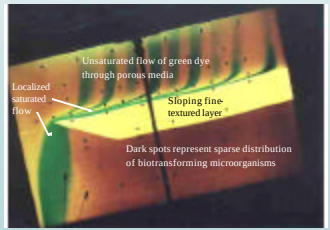
2. Inject methane, nitrous oxide, trichlorophosphate as gaseous C, N, P for indigenous microorganisms.



Research Status (Project initiated fall, 1999)

1. Nearest term opportunity for Hanford deep vadose zone CCl_4 sampling appears to be with the Innovative Treatment Remediation Demonstration (ITRD) Program, sometime in 2000.
2. Laboratory studies with injection of organisms & nutrient solutes
   - Proposal was focused only on gas-phase nutrient injection.
   - However, subsequent discussions with DOE-Richland identified injection of non-engineered microorganisms & aqueous-based nutrients as potentially acceptable technologies at DOE sites.
   - Thus, initial experiments are being performed with *Pseudomonas stutzeri* strain KC. This strain degrades CCl_4 rapidly with little to no production of chloroform and has been shown to be effective in a saturated zone CCl_4 bioremediation field demonstration in Michigan.
3. Experimental & modeling research of unsaturated zone microbial processes
   - Building off National Science Foundation (NSF) funding to Dr. Selker.
   - Transitioning from organism & substrate used in NSF studies to DOE-relevant organisms & substrates.
   - Construction of green fluorescent protein -reporter organisms for real-time monitoring in vadose visualization chambers.
4. Modeling of the impact of heterogeneity on bioremediation.
   - Previous research support from DOE EMSP and Subsurface Science Program funding
   - Completion of modeling and manuscript writing with funding from this project

Chamber for Studying Effects of Physical & Biological Heterogeneities on Unsaturated Zone Contaminant Fate & Transport



(image from Oregon State Univ. NSF project)

R&D Transition Strategy

Means

- Communication with companies managing Hanford Site clean-up
- Communication to other interested parties (fred.brockman@pnl.gov)

Useful information

1) Feasibility of bioremediation of CCl_4 in the Hanford vadose zone
   - 1st priority = point injections of nutrient solutes and organisms
   - Gas-phase nutrient injection (no water or organism additions)
   - Efficacy of microbial colonization of the vadose zone and the physical & hydrologic factors controlling the process is critical to both approaches
2) A vadose zone reactive transport model incorporating microbial processes
   - Incorporation of microbial processes into STOMP (includes prior NSF work)
   - Validate and refine model with emperical laboratory data
   - Focus on hydrologic & microbiological heterogeneities and interactions
3) Guidelines on the level of site characterization needed for accurate modeling of engineered vadose zone bioremediation