**Project Summary, No. 54681: Dynamics of Coupled Microbial and Contaminant Transport**.

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**Number of graduate students involved:** 3 (UC Davis), **Number of postdocs:** 1 (Purdue), 1 (U S. Carolina).

**Specific DOE problems being addressed/potential practical relevance:** The general problem addressed is the subsurface contamination by chlorinated solvents, and the remediation of chlorinated hydrocarbon contamination by anaerobic consortium involving microbiological agents that undergo transport and growth. This project has potential practical relevance to improved knowledge about how subsurface anaerobes interact dynamically *in situ*, and about how we can control subsurface conditions in order to manipulate the transport properties of microbes, thus allowing controlled mobility to and throughout contaminated regions, and/or controlled immobilization in the preparation of *in situ* biobarriers.

**Research Objective:** Dynamic microbial attachment/detachment occurs in subsurface systems in response to changing environmental conditions caused by contaminant movement and degradation. This project's objective is to develop the understanding of the environmental conditions and mechanisms by which anaerobic bacteria partition between aqueous and solid phases. In particular this interdisciplinary research project provides fundamental information on the attachment/detachment dynamics of anaerobic bacteria in heterogeneous porous media under growth and growth-limiting conditions. This is a critical requirement for designing and evaluating in situ bioremediation efforts.

**Research Progress and Implications:** This large-scale multidisciplinary project has made advances in basic and applied sciences on a number of research fronts relevant to the coupled subsurface fate and transport of chlorinated hydrocarbons and the anaerobic microbes involved in their remediation. Progress is reported from the smallest scale upwards, beginning with reaction kinetics modeling for our consortium, and attachment/detachment experiments in parallel, and through our results on reactive transport in natural media, to large scale field remediation results.

The mechanisms controlling degradation by a three-member consortium of anaerobic dechlorinating microorganisms (involving *Desulfomonile tiedjei*, *Desulfovibrio* (G-11), and *Syntrophobacter wolfei*) were unified and formulated into a mathematical model of the complete reaction system (Chilakapati et al, 1998) and that is fundamentally based on reaction energetics (Brugato, 2000). Explicit accounting of reaction energies allows for consistent treatment of endproduct (hydrogen) inhibition of growth and degradation, as well as an accounting of maintenance energy requirements that is crucial for our consortium.

The interaction of microbes in this consortium affects their transport behaviour in ways previously unknown; experiments at the University of S. Carolina utilizing both batch and microflow chamber (http://www.baruch.sc.edu/robb/) conditions have been completed that identify important differences between transport properties of consortium members, as well as significant dependencies of these properties on microbial growth (Van Schie and Fletcher, 1999). Measurements of transport of cells through hydrophobic-interaction and electro-static-interaction columns indicated that all species had negatively charged cell surfaces and that D. tiedjei and G-11 possessed some hydrophobic cell surface properties. Starvation greatly decreases adhesion of S. wolfei and Desulfovibrio sp. strain G11 but seems to have less of an effect on the adhesion of the other bacteria. The presence of Fe 31 on the substratum significantly increased the adhesion of S. wolfei, whereas the presence of silicon hydrophobic groups decreased the numbers of attached cells of all species. Preliminary modeling of these effects is reported in Smits and Cushman (1999a, 1999b).

The interactions of the contaminant with the degrading culture *in situ* depends greatly on the transport processes in the subsurface that control the availability and residence-time of contaminant as substrate for consortium activity and growth (Murphy and Ginn, 2000). These processes include diffusive mass transfer of solute contaminant into low-conductivity regions (Basagaoglu et al., 1999), that commonly defeats remediation strategies that rely on pumping to remove contaminants for biobarrier or *ex situ* remediation. Basagaoglu et al (1999) develop a new simplified modeling approach for keeping track of solute diffusion effects in such media, as it affects the availability of solute hydrocarbon for anaerobic degradation. Field-scale transport in the subsurface involves uncharacterized heterogeneities in physical and chemical properties governing flow and transport (Cushman and Hu, 1997; Ginn, 1998; LaViolette et al., 1999; Cushman and Ginn, 2000), and upscaling the biodegradation processes here requires careful treatment of the dynamic couplings between flow, transport, growth, and bacterial adhesion; a field-scale modeling framework for this is provided through streamtube-ensemble modeling approach (Ginn, 2000) that approximates transport in the field via use of a series of streamtubes. The streamtube approximation allows the reduction in the dimensionality of the problem from three-dimensions to (a series of) one-dimensional effective streamtubes. It also allows the modeling to rely on realistically-available field data such as tracer test results, as opposed to requiring the complete map of the subsurface flow properties (e.g., hydraulic conductivity and porosity) which is never available. Finally, this approach is extended for applicability to our consortium by keeping track of the complete reaction network describing growth, degradation, and bacterial adhesion (Murphy and Ginn, 2000).

A brief summary of implications determined under this research project is as follows: anaerobe transport properties vary in a describable fashion with activity level and degradation; S. Wolfeii in particular increases its mobility dramatically under oligotrophic conditions; D. tiedjeii generally forms a biofilm under all conditions studied; the presence of D. tiedjeii increases the attachment of other consortium members. Anaerobe degradation rates are in fact affected by endproduct inhibition and maintenance requirements under commonly encountered subsurface conditions, and a model of these effects is developed. The combined effects of growth-couplings to bacterial transport, and solute substrate fate and transport in natural systems has dramatic implications for bioremediation planning and natural attenuation. In particular, diffusive mass transport into low-conductivity zones limits the availability of solute to S. Wolfeii which under starving conditions will decrease its attachment rate
and be transported away by convection; amending the groundwater with alternative electron-acceptors and/or with other microbial species can ameliorate this problem. To complement these results, models have been developed for the reaction kinetics, for the development of biofilms and other residence-time effects on bacterial adhesion, for transport in heterogeneous flow fields, and for complete fate (e.g., reaction systems) and transport in the field as arises in *in situ* remediation planning and simulation.

**Planned Activities:**

The project is continuing to the end of the 2000 calendar year via no-cost extensions, and will complete the final experiments allowing exercise and validation of the streamtube-ensemble modeling incorporating both the biodegradation kinetics and the bacterial adhesion interactions identified among members of our consortium. As part of the experimental planning, a large intermediate-scale flow chamber was constructed and is available at PNNL. The intermediate-scale experiments will build on column experiments already conducted by Dr. Murphy at PNNL that show consistent behaviour of the consortium member adhesion dependencies on growth as previously identified in Dr. Fletcher's laboratory -the modeling and manuscript preparation from these column experiments is underway, and the modeling and reporting of the intermediate-scale experiment will conclude project work at the end of the calendar year.

**Information Access - Web Pages:**

**http://www.baruch.sc.edu/robb/** details construction of and experiments involving microflow chamber for observing anaerobe adhesion under different growth conditions.

**http://www.math.purdue.edu/~jcushman/jcushman.html#dispersion** summarizes continuing research on contaminant transport behaviour in natural subsurface environments.

**Information Access - Publications - Papers**

Basagaoglu, H., T. R. Ginn, B. J. McCoy, M. A. Marino, A linear driving force approximation to a radial diffusive model, submitted, *Am. Inst. Chem. Eng. J.*, December 1999.

Brugato, C. J., Mathematical Modeling of an Anaerobic Syntrophic Butyrate-Degrading Coculture, Master of Science Thesis, Department of Civil and Environmental Engineering, University of CA, Davis, March, 2000.

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Cushman, J. H. and T. R. Ginn, The fractional ADE is a classical mass balance with convoution-fickian flux, submitted, *Water Resources Research*, February 2000.

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Emerick, R. W., F. J. Loge, T. R. Ginn, and J. L. Darby. Modeling the Inactivation of coliform bacteria associated with particles, submitted, *Water Env. Res*., August 1999.

Ginn, T. R., Streamtube-ensemble techniques for nonlinear multicomponent reactive transport in heterogeneous media, in R. Govindaraju (ed.) Stochastic Methods in Subsurface Contaminant Hydrology, ASCE, in press, 2000.

Ginn, T.R. Comment on "Stochastic analysis of oxygen-limited biodegradation in three dimensionally heterogeneous aquifers," by F. Miralles-Wilhelm, L. W. Gelhar, and V. Kapoor, *Water Resources Research*, 34(9):2423-2426, 1998.

LaViolette, R. A., M. E. Watwood, T. R. Ginn, and D. L. Stoner, Spatial disorder and degradation kinetics in intrinsic biodegradation schemes, *J. Physical Chem A*, 103(23): 4480-4484, 1999.

Moroni, M., and J. C. Cushman, Using Statistical Mechanical Theories in Conjunction with 3D-PTV Experiments to Study Pre-asymptotic Dispersion of a Conservative Tracer in Porous Media, submitted, *Water Resources Research*, September 1999.

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Murphy, E. M. and T. R. Ginn. Modeling microbial processes in porous media, , *Hydrogeology J*., vol. 8 no. 1, pages. 142-158, 2000.

Smits , R.G. and J. H. Cushman, A Random Walk Model for Microbial Dynamics in Porous Media, Part I, submitted July 1999a, *Chem. Eng. Sci.*.

Smits , R.G. and J. H. Cushman, A Random Walk Model for Microbial Dynamics in Porous Media, Part II, submitted July 1999b, *Chem. Eng. Sci.*.

Van Schie, P. M. and M. Fletcher, Adhesion of Biodegradative Anaerobic Bacteria to Solid Surfaces, *Applied and Environmental Microbiology*, Vol. 65, No. 11, p. 5082–5088, Nov. 1999.

**Information Access - Publications - Abstracts/Posters Selection (only a subset shown for space savings):**

Murphy, E. M., B. D. Wood, T. R. Ginn, J. H. Cushman, R. Smits, Y. Gorby, M. F. Romine, K. B. Wagnon, 1998. Tracking the Transport of Dissimilatory Iron-Reducing Bacteria in Porous Media by Confocal Microscopy, AGU Fall Meeting, San Francisco, CA, December, 1998, EOS Trans., 79(45):F325.

Ginn, T. R., T. D. Scheibe, E. M. Murphy, M. F. DeFlaun, T. C. Onstott, 1998, Effects of chemical heterogeneity on subsurface fate and transport involving biotic reaction systems: Two Examples. AGU Fall Meeting, San Francisco, CA, December, 1998, EOS Trans., 79(45):F294.

Murphy, E. M., T. R. Ginn, F. J. Brockman, and D. R. Boone, 1997. Growth Effects on the Partitioning and Transport of Bacteria, AGU Fall Meeting, San Francisco, CA, Dec. 1997, EOS Trans., Vol 78 (46), pp F231.

Van Schie, P.M. , M. Fletcher, "Characterization of the adhesive behavior of anaerobic bacteria with bioremediation potential". Poster presented at the American Society for Microbiology Annual Meeting, May 30-June 3, 1999, Chicago, Illinois.

Van Schie, P.M. , D.R. Boone, M. Fletcher, "Adhesion of biodegradative anerobic bacteria to solid surfaces." Poster presented at the American Society for Microbiology Annual Meeting, May 17-21,1998, Atlanta, GA.