In Situ Bioreactor: A New Tool to Help Improve Biostimulation and Bioaugmentation

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Background/Objectives. The remedial amendments and bioaugmentation cultures commercially available today are the result of considerable research and development. However, there's not a universal approach to enhanced biodegradation as each site presents its own unique challenges. For example, even after a successful amendment injection, elevated contaminant concentrations could inhibit contaminant-degrading microorganisms or low contaminant concentrations may not be able to sustain an active population of degraders. Bioaugmentation cultures enriched in the laboratory may have survivability issues in the field, and there are still many contaminants that are not addressed by commercial cultures. Three case studies will be presented where in situ bioreactors (ISBRs) built on existing Bio-Sep® bead technology have been shown to address these issues.

Approach/Activities. The ISBRs are designed to fit within a 2-inch monitoring well and can be adapted for aerobic and anaerobic treatment applications. The ISBRs are filled with Bio-Sep® beads, which provide a matrix of powdered activated carbon (PAC) and Nomex® that can be rapidly colonized by the active portion of a microbial community. The PAC adsorbs contaminants and nutrients present in the aquifer and serves to concentrate indigenous degraders for treatment purposes. Groundwater flow through the bioreactor allows for microorganisms from within the bioreactor to migrate into the formation beyond the wellbore area to further promote biodegradation in the aquifer. Amendments can be delivered into the unit via topside equipment.

ISBRs have been used to treat both petroleum hydrocarbons and chlorinated solvents as well as other contaminants of concern. This presentation will provide data from ISBR applications at an industrial site with toluene concentrations exceeding 200 mg/L, a heating oil release with low, yet persistent BTEX concentrations (<100 µg/L), and a chlorinated solvent site where an ISBR was seeded with indigenous microorganisms from a well with a robust population of *Dehalococcoides* and deployed in another area of the site where reductive dechlorination was limited.

Results/Lessons Learned. At the industrial site, an aerobic ISBR was installed in a well with very high and likely inhibitory toluene concentrations. The toluene concentration was reduced by 87% after two months of ISBR operation, and the well was in compliance by the end of the study. It is proposed that the adsorptive surface of the PAC reduced the toluene within the beads to concentrations that reduced or eliminated inhibition and stimulated biodegradation. Stable isotope probing was used to confirm that toluene biodegradation was occurring on the PAC. The aerobic ISBR installed at the heating oil release successfully stimulated biodegradation as indicated by a decrease in petroleum hydrocarbons and a concurrent increase in expression of genes associated with aerobic hydrocarbon biodegradation. This degree of stimulation was not produced by air sparging and nutrient addition alone. Finally, at the chlorinated solvent site, *Dehalococcoides* was maintained and daughter products were detected at the well where the anaerobic ISBR was deployed.