Validation of Anaerobic Benzene Bioaugmentation Approaches through Bench-Scale Treatability Studies

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Background/Objectives. Biodegradation of benzene, toluene, ethylbenzene and xylene (BTEX) by microorganisms is a significant process that is known to occur in diverse environments. Understanding this process has important implications for the application of bioremediation technologies at BTEX-contaminated sites. While aerobic microbial processes can degrade BTEX more rapidly than anaerobic processes, aerobic bioremediation is not feasible at all sites (i.e., deep anaerobic aquifer systems where oxygen addition is prohibitive). In these instances, anaerobic approaches can be used to better address BTEX contamination.

Over the past 20 years, anaerobic (methanogenic) enrichment cultures capable of complete degradation of benzene, toluene and *o*-xylene to carbon dioxide and methane have been developed at the University of Toronto. The cultures have recently been characterized using next-generation DNA sequencing technologies and other genomic tools that have helped identify the key organisms associated with benzene, toluene and xylene degradation (Luo et al., 2015).

Approach/Activities. SiREM and researchers at the University of Toronto have collaborated to scale up an anaerobic benzene bioaugmentation culture from research to commercial volumes through a 3-year research project. The culture has been used in multiple laboratory treatability studies to evaluate its performance to remediate a variety of hydrocarbon-contaminated materials from field sites in the US, Canada, China and Germany. Degradation of BTEX was monitored with and without bioaugmentation and under varying electron acceptor conditions. The growth of the benzene-degrading microbes was tracked by quantitative polymerase chain reactions to establish correspondence between the growth of these organisms and benzene degradation. Information developed from this testing includes degradation rates and the range of geochemical conditions required for optimal performance, which are being used to design field pilot trials. These laboratory studies have also been used to validate molecular biomarkers used to monitor in situ biodegradation activity.

Results/Lessons Learned. To date, benzene bioremediation potential has been reported in nine of ten site materials surveyed, demonstrating that anaerobic biodegradation is a feasible benzene clean up strategy for field applications. At one site, bioaugmentation increased rates of benzene degradation by an average of 13- and 37-fold as compared to nitrate biostimulation and natural attenuation, respectively. Successful culture scale-up, treatability studies and molecular tools have laid the foundation for upcoming field pilot tests.