Enhanced Control of Biomass Production and Microbial Activity via Acetylene Inhibition during TCE Aerobic Cometabolism

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Background/Objectives. Bioremediation of large diffuse contaminant plumes often experience challenges such as limited substrate transport and excessive microbial growth near substrate injection points leading to bioclogging. Bioclogging decreases permeability and effectiveness of in situ bioremediation. In this project, we are investigating the effect of acetylene, a microbial inhibitor, on cometabolic aerobic biodegradation of trichloroethene (TCE) using propane and oxygen as growth substrates. Bioremediation of TCE via aerobic cometabolism is a potentially feasible approach for dilute plumes. In order for aerobic cometabolism to occur, microbes must be provided growth substrates. We provided propane gas and oxygen. Microbes biodegrade propane and an initial enzyme in the biodegradation process is a monooxygenase, which inserts one \(-\text{O}\) or \(-\text{OH}\) group into the substrate. Monooxygenases degrade both the growth substrate and cometabolically degrade TCE. As a microbial inhibitor, acetylene binds to monooxygenase enzymes inactivating them for propane metabolism and TCE cometabolism. Microbes overcome this inhibition by producing new copies of the monooxygenase enzyme. This highly specific microbial inhibition may allow bioremediation practitioners the capacity to slow in situ biomass production, reduce bioclogging and create larger, more homogenous zones of in situ contaminant remediation.

Approach/Activities. Batch studies were conducted using two microbial cultures: a pure culture of the hydrocarbon-degrading Mycobacterium austroafricanum JOB5 and a hydrocarbon-degrading mixed culture enriched from soil. These cultures were fed with propane and oxygen and were exposed to TCE (50 µM) to verify aerobic cometabolic capacity. They were then exposed to monooxygenase-inhibiting acetylene gas at 5% v/v (in the headspace) for differing lengths of time (no exposure (0 day), 1 day, 2 days, 4 days, and 8 days) to assess how microbial growth, substrate consumption, and TCE cometabolic capacities were altered.

Results/Lessons Learned. In batch incubations, both cultures aerobically cometabolized TCE and exhibited time-dependent relationships between acetylene exposure and microbial growth, and growth substrate consumption. TCE cometabolism rates and substrate consumption decreased when the length of acetylene exposure was greater than 2 days. Biomass production was lower in the incubations with acetylene. Without acetylene exposure, M. austroafricanum JOB5 and the mixed culture completely degraded the TCE concentration provided within 45 days at an average rate of 0.76 and 1.76 µmol TCE L\(^{-1}\) d\(^{-1}\), respectively. Exposing the cultures to 8 days of acetylene decreased the cometabolic rate by 7-fold in the mixed culture and by 11-fold and by in the M. austroafricanum JOB5 microcosms with 20 µM TCE still remaining in both cultures at day 60 of incubation. The mixed culture produced less biomass than M. austroafricanum JOB5, even though it exhibited higher TCE cometabolic rates.