Effect of Nitrate Stress on Metal-reducing Microbes and Results of Nitrate push/pull Field Tests at Hanford 100H.

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Project Goal: The ENIGMA (Ecosystems and Networks Integrated with Genes and Molecular Assemblies) project seeks to elucidate the mechanisms and key processes that enable microorganisms and their communities to function in metal-contaminated soil sites. One goal is to understand the effect of environmental stressors that enable ENIGMA-relevant microorganisms to thrive in such environments.

Abstract: As part of the ongoing investigation of sustainable bioremediation of Cr(VI) in groundwater at the Hanford 100H area, we performed groundwater biostimulation tests by injecting Hydrogen Release Compound (HRC) and three lactate (17mM) injection experiments. To investigate the response of resident microbes to nitrate stress, a push-pull test was then conducted by injecting 55 gals of groundwater (collected from the background well) with KNO_3 (nitrate concentration 5,000 ppm) in October, 2010. After one day, pumping began from the same well, and lasted for 16 days. As a result of nitrate injection, total biomass decreased and sulfate concentration increased, but the sulfide and iron concentrations dropped. During pumping, the nitrate concentration decreased about 3 orders of magnitude. PLFA data showed biomass on the order of 10^7 cells/ml prior to push pull, and dropping off to 10^5 cells/ml during the test, but recovering toward the end back to 10^7 cells/ml by the end. Biomarker lipids indicate a shift toward monoenoics indicating an increase in gram negative bacteria and decrease in branched lipids (gram postive) and branched monoenoic (sulfate reducers).
We discuss the field test results in details and elucidate the effect of nitrate stress on environmentally relevant microbes *Geobacter metallireducens* and *Desulfovibrio vulgaris* as observed through controlled lab experiments. The lab studies and the field study together will help in understanding the overall fate of microbes under changing environmental conditions in the field and the key cellular mechanisms impacted by such stress conditions.

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