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Measuring Biotic Soil Hydrogen Demand as a Strategy for Bioremediation Potential Assessment

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Background/Objectives. Bioremediation of chlorinated solvents via bioaugmentation is an effective strategy to remedy contaminated sites. However, it is difficult to predict the extent of success of bioaugmentation from one site to another. We hypothesize that success or failure at bioaugmentation is due largely to the competition *Dehalococcoides* faces with other hydrogen oxidizing microbes. Competition can be quantified by measuring a total biological hydrogen demand (TBHD), or the sum of all electron acceptors that can be used by hydrogen oxidizing microbes, of site specific soil. Our aim was to track hydrogen consumed in the subsurface and establish a recommended protocol to address biological hydrogen demand needs to allow for more accurate assessments of bioremediation potential at contaminated sites.

Approach/Activities. Biotic soil hydrogen demand was measured in soils from fifteen separate sites. Saturated, anaerobic microcosms were made for each soil with hydrogen as the sole electron donor and soil associated ions as the sole electron acceptors. Microcosms were repeatedly spiked with hydrogen after each previous dose was consumed for up to 200 days. Hydrogen consumption was measured by subtraction with gas chromatography. Metabolic products like volatile fatty acids were measured with high throughput liquid chromatography, and soil associated electron acceptors were measured using ion chromatography. DNA from the soil samples was extracted at the beginning and end of the experiment and 16S rRNA gene amplicons were sequenced with Illumina Miseq technology.

Results/Lessons Learned. We completed an electron balance on the added hydrogen and tracked its partitioning to different microbial processes. The results showed that reduction of soil associated electron acceptors is a significant part of the hydrogen flux in the subsurface. The soil geochemistry plays a large role in hydrogen partitioning to different microbial processes and strongly influences the resulting microbial community structure and function. In soils with high concentrations of soil associated electron acceptors like nitrate and sulfate, added hydrogen was used for reduction of these compounds. In soils with low concentrations of soil associated electron acceptors, added hydrogen was instead used to produce organic compounds like acetate. These results highlight the influence of soil geochemistry on the fate of hydrogen in the subsurface and inform how microbial communities and soils might be managed to direct the flow of hydrogen to *Dehalococcoides* and improve remediation success in the future.