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PHYLOGENETIC AND FUNCTIONAL GENE MICROARRAY ANALYSIS DEMONSTRATES DIRECT AND INDIRECT MECHANISMS FOR SUSTAINED CHROMIUM BIO-IMMOBILIZATION.

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Background: During a field-scale chromium treatability study, a single dose of a slow release electron donor (HRC) was applied to a contaminated aquifer to stimulate microbial reductive precipitation of hexavalent chromium Cr(VI). Here we present analysis of microarray-based prokaryotic population dynamics and correlations with geochemical and geophysical observations following this application over two years. Methods: A high-density 16S rRNA phylogenetic microarray (PhyloChip) and a functional gene array (FGA) were used to analyze groundwater samples from multiple depths in injection and monitoring wells taken at intervals pre- and post-HRC injection. Following filtration, genomic DNA was extracted and PCR amplicons or MDAamplified community-DNA were analyzed by microarray hybridization. A range of geochemical and geophysical parameters were also monitored. Results: Following HRC injection reducing conditions had rapidly established with a corresponding decline in DO, Eh and nitrate. Cr(VI) concentrations declined steadily over 6 weeks and remained below upgradient concentrations. PhyloChip data demonstrated depth stratified microbial communities with temporal shifts in composition corresponding with observed geochemistry. A sustained enrichment of iron and sulfate reducing bacteria was observed over 2 years, suggesting indirect chromium immobilization through interaction with reactive iron or sulfide by-products. Nitrate reducers such as Pseudomonas also remained elevated over the two years and FGA array data demonstrated a sustained enrichment of Pseudomonas chromate reductase genes suggesting direct reduction of chromate may also be significant in chromium immobilization. Based on this data, organisms representing each of these functional groups have been isolated and characterized. Conclusions: The combination of phylogenetic and functional gene arrays represents a complementary highthroughput approach to elucidating mechanisms responsible for contaminant immobilization in the subsurface.