Coupled Chemical Pre-oxidation and Aerobic Biodegradation of Buried MC252 Oil across a Headlands Beach Profile

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Background/Objectives. Hard structures used on Fourchon Beach, Louisiana to mitigate the transport of MC252 oil into adjacent marshes established conditions for oil burial beneath the groundwater table across the beach profile. Persistence of 3- and 4-ring PAHs and n-alkanes has been observed in subsurface sediments at the field site. Groundwater is hypersaline (>50 ppt), anaerobic (DO well below 0.1 mg/L) and redox conditions are reducing (ORP<-200 mV). Oxygen flux to the subsurface oil deposit is limited, impeding the natural biodegradation of crude oil. Coupled in situ oxidation processes chemical pre-oxidation with activated persulfate and enhanced bioremediation through the use of an oxygen releasing compound were used to stimulate biodegradation of buried petroleum hydrocarbons.

Approach/Activities. Two treatment application phases were employed at the site: Phase 1 (P1-02/2018) consisted of initial application of persulfate oxidant within the approximately 60 m2 treatment zone and Phase 2 (P2-11/2018) incorporated the subsequent addition of persulfate with a co-application of an oxygen release compound. Distribution of PAHs and n-alkane compounds pre- and post- treatment were surveyed by sediment extraction and GC-MS. The impacts of treatment phases on groundwater chemistry were continually monitored (ORP, pH, conductivity, temperature, and DO). Bacterial communities were characterized in samples from before and after P1 (02/2018-05/2018) using Illumina Miseq profiles of PCR-amplified 16S rRNA gene fragments. To investigate changes encountered within microbial communities, NMDS and AMOVA analyses were completed along with LEfSe analysis to describe which populations were responsible for significant differences between samples from different dates.

Results/Lessons Learned. Chemical pre-oxidation with persulfate degraded a differential portion of target contaminants through a variety of non-specific oxidation reactions. Significant removal of PAHs and n-alkanes for those samples classified as highly contaminated (>100 mg/kg total PAHs) and low contaminated (<15 mg/kg total PAHs) samples were observed after P1 oxidant application. In spite of changes in oxidative stress and increases in sulfate concentrations, resilience of the microbial population was observed after P1. Analysis illustrated that chemical oxidation caused significant differences in microbial community structure, however samples taken 1 month after P1 were enriched with similar genera as those from pre-P1 samples, indicating a resiliency within the population. Substantial increases in microbial diversity 3 months after P1 may indicate a resistance to the impacts of chemical oxidation, highlighting an ability for the regeneration of normal microbial function. Phylotypes belonging to the class Gammaproteobacteria were dominant for each sample date, a number of which are known hydrocarbon degraders typically associated with oil contamination. Among Gammaproteobacteria, *Marinobacter* spp. was dominant for all sampling dates however had statistically elevated 16S gene copies in pre-P1 samples. The relative abundance of Deltaproteobacteria increased from 4% to 16% 3 months after P1. The following genera were detected at significant levels 3 months after P1: *Aliidiomarina, Halanaerobium, Methylohalobius, Sulfurimonas, Thiomicrospira,* and *Thioalkalimicrobium.* Oxygen concentrations from ORC is expected to ease the environmental pressures associated with P2 application and to streamline the regeneration and capabilities of the microbial community. Results from this research can lead to improved understanding of the limitations associated with applying aerobic bioremediation strategies to oxygen-limited environments.