Use of Molecular Biological Tools and CSIA to Assess Natural Attenuation of MTBE and TBA

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Background/Objectives. Failure of a storage tank fill line resulted in multiple releases containing pure phase MTBE and gasoline range hydrocarbons. Current dissolved MTBE and TBA concentrations range up to 2,500,000 µg/L and 140,000 µg/L, respectively. A study was conducted to evaluate natural attenuation of MTBE and TBA. Objectives consisted of assessing whether the presence of TBA was related to transformation of MTBE or dissolution from a gasoline blend, characterizing the microbial community for MTBE and TBA degraders, identifying the dominant degradation pathways, and utilizing chemical, isotopic and microbial analysis to assess the magnitude and extent of MTBE and TBA degradation in different representative areas of the site consisting of the plume core, plume core periphery, and areas downgradient of the plume core including a tidally influenced marsh.

Approach/Activities. Equilibrium partitioning analysis was used to assess whether the presence of TBA is due to dissolution or biotransformation of MTBE. Soil samples were collected from five soil borings and sent to a laboratory for phospholipid fatty acid (PLFA) analysis and quantitative polymerase chain reaction (qPCR) analysis. Groundwater samples were collected from 15 monitoring wells and sent to a laboratory for chemical analysis including TPH-gas, BTEX, fuel oxygenates MTBE and TBA, and geochemical parameters, microbial analysis including PLFA and qPCR analysis, and compound specific isotope analysis (CSIA) of carbon and hydrogen associated with both MTBE and TBA. Six advanced Bio-Trap[®] samplers loaded with ¹³C-labeled MTBE and three Bio-Trap[®] samplers loaded with ¹³C-labeled TBA were deployed in groundwater monitoring wells for at least 30 days and sent to a laboratory for qPCR, PLFA, and stable isotope probing (SIP) analysis.

Results/Lessons Learned. Increasing TBA/MTBE ratios and equilibrium partitioning analysis suggest the presence of TBA in the plume core periphery and downgradient areas is primarily a result of MTBE degradation, whereas TBA in the plume core is more likely due to dissolution from MTBE product. Geochemical parameters are indicative of methanogenic conditions in the plume core, metal and sulfate reducing conditions in the plume core periphery, and a mix of sulfate reducing and oxic conditions in the tidally influenced marsh. gPCR and PLFA results provide evidence of anaerobic degraders in the plume core where anoxic conditions persist and both anaerobic and aerobic degraders in the marsh. Enrichment of MTBE ¹³C and ²H isotopes increased as a function of distance from the release area, with stronger enrichment observed in the ¹³C isotope suggesting anaerobic degradation is the dominant pathway. Enrichment in TBA ¹³C and MTBE ²H and a corresponding depletion in TBA ²H was observed in marsh monitoring wells, which is evidence of aerobic degradation of both MTBE and TBA. Bio-trap® SIP results showed MTBE and TBA isotope-labelled mass loss in all samples, with the highest levels of enrichment in biomass, PLFA, and dissolved CO₂ in plume core periphery and downgradient wells. Attenuation rate constants were highest in downgradient wells, including wells in the marsh which exhibited half-life's ranging from 1.1 to 7.2 years. Overall, multiple lines of evidence demonstrate MTBE and TBA degradation on the periphery of the plume core and in the tidally influenced marsh and little to no biodegradation in the plume core. Results suggest active remediation of the plume core will be required to stimulate intrinsic biodegradation, while existing downgradient conditions in the marsh appear to be attenuating MTBE and TBA.