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Naphthalene Stable Isotope Probing Illustrates That Sulfate Amendment Enhances Biodegradation at a Former Manufactured Gas Plant

Monica Heintz (monica.heintz@arcadis.com) (Arcadis U.S., Inc., Denver, CO, USA)
Jeff McDonough (Arcadis U.S., Inc., Newtown, PA, USA)
John Brussel, Catherine Geraci, and Matt Hysell (Arcadis U.S., Inc., Syracuse, NY, USA)
James Morgan (National Grid, Syracuse, NY, USA)

Background/Objectives. A naphthalene stable isotope probing (SIP) study using Bio-Trap® samplers was conducted at a former manufactured gas plant (MGP) site in Upstate New York to assess the feasibility of enhancing biodegradation of MGP-related constituents with sulfate amendment. Anaerobic biological oxidation (ABOx) using sulfate as a terminal electron acceptor was selected for field-scale testing based on bench-scale testing and intrinsic geochemistry. Naphthalene-baited SIP Bio-Traps® contain Bio-Sep® beads that are impregnated with a low and quantified concentration of naphthalene with a carbon-13 isotope label (^{13}C-naphthalene). This label is strongly ^{13}C-enriched relative to natural background, and therefore carbon atoms with this label can be tracked. Thus, biodegradation of ^{13}C-naphthalene is evident based on a decrease in the ^{13}C-naphthalene content, and quantification of ^{13}C-enriched biodegradation products (^{13}C-biomass, ^{13}C-dissolved inorganic carbon [DIC], and ^{13}C-methane). SIP studies are a particularly valuable tool as they provide a definitive line of evidence for biodegradation. Additionally, the microbial community that colonizes Bio-Trap® samplers can be characterized based on biological molecules including phospholipid fatty acids (PLFA) and deoxyribonucleic acid (DNA).

Approach/Activities. Naphthalene-baited SIP Bio-Traps® were deployed at three monitoring wells during two phases: baseline and treatment. This approach enabled a direct comparison of ambient and engineered sulfate concentrations on the biodegradation of naphthalene. During the treatment phase, approximately 6,500 gallons of a solution containing 20,000 mg/L sulfate and a fluorescein dye tracer was introduced into an injection well located approximately 15 to 25 feet hydraulically upgradient of monitoring wells where Bio-Traps® were deployed. Upon Bio-Trap® retrieval, concentrations of ^{13}C-naphthalene, ^{13}C-biomass, ^{13}C-DIC, and ^{13}C-methane were measured, and PLFA and DNA were extracted. The PLFA profile was determined, and quantitative polymerase chain reaction (qPCR) analysis was applied to enumerate key genes for assessing microbial community composition.

Results/Lessons Learned. The addition of sulfate enhanced biodegradation of naphthalene. This was demonstrated by greater ^{13}C-naphthalene loss from Bio-Trap® samplers and greater incorporation of ^{13}C into DIC and biomass in the samplers deployed during the treatment phase compared to the baseline phase. The addition of sulfate also resulted in a change in the microbial community, with increased populations of sulfate reducing bacteria. Although naphthalene degradation was stimulated during the treatment-phase, the naphthalene carboxylase gene was not detected. This observation indicates that site-specific naphthalene degradation may be mediated by enzymes that are not encoded by the particular gene target commercially available. This study resulted in agency approval of ABOx for MGP-related constituents. The demonstration approach used here is applicable to other former-MGP facilities, and may also be modified to test a wide range of amendments for their potential to stimulate biodegradation.