# In Situ Remediation of a 1,4-Dioxane Plume in a Heterogeneous Aquifer, Pilot Study ISB with Bioaugmentation

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**Background/Objectives.** The goal is active remediation of a plume of 1,4-dioxane (40 to 70 feet below ground surface) in a heterogeneous aquifer in the Georgetown neighborhood of Seattle, Washington. This aquifer consists of discontinuous interbedded fine sand, silty sand, and sandy silt lenses with an estimated linear groundwater velocity of approximately 90 feet per year. The conceptual model for 1,4-dioxane transport is slow release from the low permeability units within the aquifer, which are acting as a secondary source (no primary source of 1,4-dioxane remains).

In situ biodegradation (ISB) was bench and pilot tested as an alternative to more traditional 1,4-dioxane treatment methods by in situ chemical oxidation (ISCO). Distributing treatment in the low permeability units and getting oxidant to persist long enough to significantly reduce the mass of 1,4-dioxane was a barrier to treatment at this site. The possibility of microorganisms capable of degrading 1,4-dioxane in situ has been presented in several papers reopening bioremediation as a treatment alternative in the form as passive flow through bio-barriers.

**Approach/Activities.** ISB pilot testing included a laboratory based microcosm study to isolate indigenous 1,4-dioxane degrading microorganisms and assess bioaugmentation of the aquifer. The microcosm study was completed in 2016 and did not identify statistically significant evidence pointing to indigenous organisms capable of degrading 1,4-dioxane. However, the microcosms bioaugmented with *Pseudonocardia dioxanivorans* CB1190 and *Mycobacterium* sp. PH-06 displayed robust 1,4-dioxane degradation. Additional studies were performed on low oxygen performance and performance with oxygen release compounds.

In situ pilot study of bioaugmentation was conducted in 2017 with genetic analysis concluding in February 2018. In situ bioaugmentation testing was performed at two different wells (a high concentration well [700 µg/L] and medium concentration well [200 ug/L]) and consisted of injecting a total of $1.1 \times 10^{13}$ cells of an artificial microbial consortium (SEN001) in approximately 600 mL AMS media. Under batch conditions, SEN001 demonstrated the capacity to degrade 1,4-dioxane from 500 ppm to less than 25 ppb. Groundwater monitoring (conducted for approximately 1 year) included field parameters (DO, pH, ORP, temperature, and specific conductance), standard laboratory analysis (1,4-dioxane and dissolved iron) as well as genetic analysis. Genetic analysis was performed by Sentinel Environmental and included Illumina 16S rRNA sequencing at 20,000 sequences per sample to provide genus-level microbial community structure.

**Results/Lessons Learned.** No substantial 1,4-dioxane removal was observed in either well over a six-month period. Post-bioaugmentation, the microbial community in both wells was initially dominated by species present in the injected culture, however indigenous species became more abundant with each sampling event. These results suggest that the performance and growth of the bioaugmentation culture (as well as indigenous dioxane metabolism) were hindered by insufficient oxygen availability.