

## *In Situ* Microcosms (ISMs)

### Screening MNA vs Sulfate Injection at a BTEX Impacted Site



#### PROJECT SUMMARY



- An *In Situ* Microcosm study was conducted to evaluate MNA and enhanced anaerobic bioremediation at a petroleum impacted site.
- ISM assemblies, each composed of an unamended MNA unit and a Sulfate Amended unit, were deployed in existing monitoring wells for 60 days.
- Following in-well deployment, CENSUS qPCR demonstrated that concentrations of anaerobic benzene carboxylase (ABC) and benzylsuccinate synthase (BSS) genes were orders of magnitude greater in the Sulfate Amended units than in the MNA units indicating that sulfate addition would stimulate growth of anaerobic BTEX degraders.

#### PROJECT CHALLENGE



Groundwater at a petroleum storage facility was impacted by benzene, toluene, ethylbenzene, and xylenes (BTEX). Subsurface conditions were highly anaerobic and site managers wanted a cost-effective way to evaluate monitored natural attenuation (MNA) and determine whether sulfate addition would stimulate growth of anaerobic BTEX degrading bacteria and enhance bioremediation.

#### IN SITU MICROCOSMS AND ANALYSIS

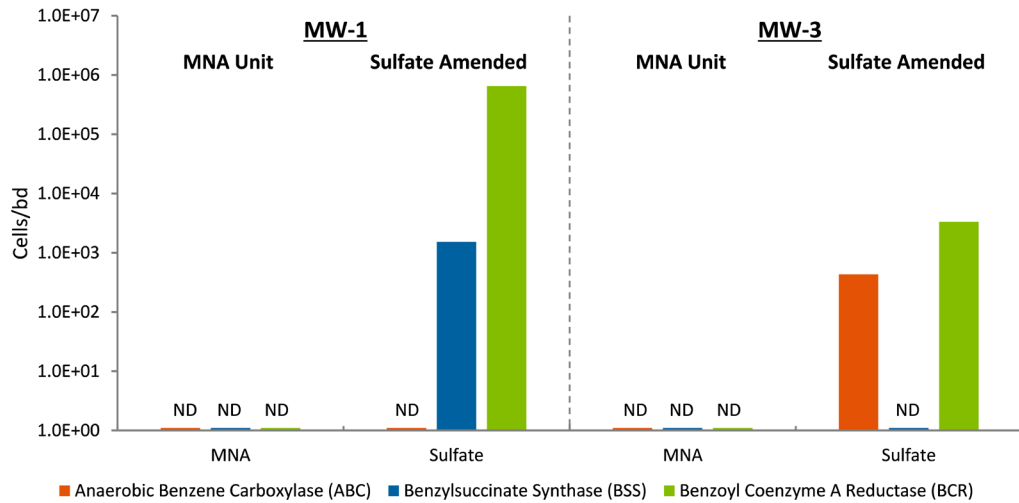


*In Situ* Microcosms (ISMs) are field deployed microcosm units, each representing a different treatment strategy. For the study site, ISMs composed of an unamended MNA unit and a Sulfate Amended unit were deployed in impacted monitoring wells for approximately 60 days. Following the in-well deployment period, the ISMs were retrieved and shipped to Microbial Insights for analysis. Each ISM unit contained passive samplers including a Bio-Trap® for CENSUS qPCR quantification of three functional genes (ABC, BSS, and BCR) involved in anaerobic BTEX biodegradation.

### ISM RESULTS



ISM assemblies with unamended MNA unit and Sulfate Amended units were deployed in monitoring wells MW-1 and MW-3. Results of post-deployment CENSUS qPCR quantification of functional genes involved in anaerobic BTEX biodegradation are shown in the figure below.



- For both MNA units, concentrations of ABC, BSS, and BCR genes were below the laboratory detection limit indicating that concentrations of anaerobic BTEX degraders were low under existing subsurface conditions.
- Conversely, functional genes involved in anaerobic BTEX biodegradation were detected in the Sulfate Amended units deployed in both monitoring wells suggesting that sulfate addition stimulated growth of anaerobic BTEX degraders.
- More specifically, BSS gene concentrations were on the order of  $10^3$  cells/bead for the Sulfate Amended unit in MW-1 while anaerobic benzene carboxylase (ABC) genes were detected at  $10^2$  cells/bead in the Sulfate Amended unit deployed in MW-3.
- Similarly, BCR genes were detected in both Sulfate Amended units but were below detection limits in the corresponding MNA units. Moreover, concentrations of BCR genes in the Sulfate Amended unit in MW-1 were relatively high, ranking at approximately the ~80<sup>th</sup> percentile in the Microbial Insights Database.

Overall, concentrations of functional genes responsible for anaerobic BTEX biodegradation were two or more orders of magnitude greater in the Sulfate Amended Units suggesting that sulfate addition would stimulate growth of known anaerobic BTEX degraders.

**Decision:** Site managers decided to inject the sulfate releasing product in source area based on historical groundwater monitoring results as well as ISM results indicating growth of anaerobic BTEX degraders in response to sulfate addition.

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## Screening MNA vs Sulfate Injection

### KEY BENEFITS



- **Cost Effective:** Simultaneous evaluation of multiple remediation options at a fraction of the cost of a lab bench treatability study or pilot scale study.
- **In Situ:** Performed in existing monitoring wells at the site under field conditions.
- **Conclusive:** Increases in three different functional genes responsible for anaerobic BTEX biodegradation in response to sulfate addition.

### LAB LOCATIONS



#### **Microbial Insights, Inc. USA**

10515 Research Drive, Knoxville, TN 37932 USA

#### **Microbial Insights Canada, c/o EBPI**

735 Griffith Court, Burlington Ontario, L7L 5R9

#### **Microbial Insights (Australia) Pty Ltd, c/o AGRF Ltd**

Plant Genomics Centre, Hartley Grove, Urrbrae SA 5064, Australia

#### **Microbial Insights Europe, c/o Avecom**

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