

Flying below the Radar: What Are Those Other Microbes Doing?

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Background/Objectives. Bioaugmentation with dechlorinating cultures is a proven technology for the remediation of chlorinated solvent contaminated sites. Specific dechlorinating bacteria and functional genes are well known to dechlorinate chlorinated solvents. *Dehalococcoides* (*Dhc*) can completely dechlorinate chlorinated ethenes to ethene, and *Dehalobacter* (*Dhb*) is involved in chloroform and 1,1,1-trichloroethane dechlorination.

Other microorganisms that have significant roles in dechlorination but are less well known and tested for less frequently; they include *Dehalogenimonas* (*Dhg*), which can dechlorinate *trans*-DCE, chlorinated propanes and more recently has been reported to dechlorinate vinyl chloride (VC) to ethene. *Geobacter* species are capable of dechlorinating high concentrations of PCE and TCE and may play a role in DNAPL dissolution as well as participating in biogeochemical reduction processes at many sites in conjunction with sulfate reducing bacteria (SRB).

Molecular biological tools are commonly used to quantify key microorganisms at contaminated sites, but typically focused on those known to degrade the contaminants of concern (i.e.; only *Dhc* analyzed at a chlorinated ethene site). Bioaugmentation cultures are mixed consortiums containing many types of microorganisms. Cultures used for chlorinated ethene sites also contain significant concentrations of *Dhb*, *Dhg*, *Geobacter* and SRB that may play important roles in remediation success.

Approach/Activities. With the number of available qPCR targets expanding and the advancement of next generation sequencing (NGS), it has become easier to identify a larger range of microorganisms in bioaugmentation cultures and at bioremediation sites that may play important roles in dechlorination. Results from qPCR and NGS analyses from both laboratory and field bioremediation applications have been collected from a wide variety of sites and analyzed with the goal of providing a more comprehensive picture of microbial process contributing to site remediation.

Results/Lessons Learned. The complexity of microbial consortia and naturally occurring microbial populations has proven to be quite vast with bioaugmentation cultures often containing several key dechlorinators and many more other microorganisms. NGS screening of sites routinely identifies several hundred-distinct species. At many sites, lesser known microbes such as *Dhg*, *Dhb*, *Geobacter*, SRB and methanogens appear to play critical roles contributing to successful bioremediation outcomes. *Dhg*, in particular, appears to be widespread and may be a major contributor to reductive dechlorination at a significant number of sites. Understanding the contributions of microbial populations important to abiotic degradation mechanisms also supports the effective use of monitored natural attenuation at sites. Key site characterization data needs, in addition to microbial characterization will also be presented.