Novel Strategies in Groundwater Bioremediation: *new solutions to old problems*

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Groundwater remediation has evolved over the last decade by blending basic research and applied engineering; contaminants that were previously considered recalcitrant are now treated using microbial activity. These contaminants include organic molecules such as explosives residues and trichloroethylene (TCE), as well as inorganic nutrients such as phosphorus. In addition, bioremediation needs to develop technologies that are more sustainable, so current cleanup activity does not adversely impact future land and water use needs. Our laboratory is developing several new approaches for groundwater bioremediation that address the contaminants above, while increasing the overall sustainability of the practice by reducing methane emissions, and utilizing waste substrates as remediation amendments.

TCE remediation has long been dominated by the concept that electron donors that are added must a) be high molecular mass compounds that ferment to hydrogen and b) be added at very high concentration to overcome "stalling" at cis-dichloroethylene (Cis-DCE). These two erroneous assumptions lead to excessively high methane production, without increasing the rate of TCE reduction to ethene, or the number of the critical microorganisms (Dehalococcoides-like cells) during the reactions. Our data demonstrate that adding acetate (the key carbon intermediate in all organic matter decomposition) at low/stoichiometric concentration actually increases the rate and extent of complete dechlorination, while generating little or no methane. Quantitative PCR demonstrated that the number of Dehalococcoides do not change as acetate increases, while the number of methanogens increases by several orders of magnitude. In addition, TCE was reduced to ethene concurrently with Fe(III) reduction, a process that has previously been characterized as inhibitory to complete dechlorination.

Explosives residues are common at military training facilities and munitions production facilities. These compounds are treated using both in situ and ex situ approaches. Our research has identified three unique ways to treat the explosive RDX by amending with what are called extracellular electron shuttles. These soluble molecules bridge microbial metabolism and RDX transformation, without the need for the cells to come in contact with the contaminant. Our data have applicability both in situ and ex situ, and recently we have incorporated photosynthetic microorganisms (Rhodobacter species) – such that sun energy can be used to degrade RDX in aboveground reactors.

Finally, the amendments that are typically used in bioremediation applications are specialty molecules synthesized from virgin source materials, with a high cost of energy and raw materials. We have identified several "waste" substrates – most notably distillers' grains (both wet and dry), and glycerol waste from biodiesel, that promote the same reaction rates as specialty bioremediation compounds. Re-using these wastes increases the overall sustainability of the processes, turns the wastes into a commodity product, and decreases the costs for site stakeholders.