Goldschmidt 2000
September 3rd–8th, 2000
Oxford, UK.

Journal of Conference Abstracts
Volume 5(2), 470

The Effect of Physical and Chemical Sediment Heterogeneity on Aerobic and Anaerobic Bacterial Transport

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Contamination of groundwater by anthropogenically derived chemicals poses a serious threat to a precious natural resource. In cases of inaccessible sites, spatially large plumes or low levels of contamination, manipulating the natural microbiological diversity occurring within the subsurface may be the only viable remedial option. One such approach is *in-situ* stimulation of micro-organisms capable of degrading the target contaminant (biostimulation). Alternatively, where such indigenous micro-organisms do not exist or are present in low numbers, bio-augmentation with appropriate exogenous or indigenous isolates may be more beneficial.

Bioaugmentation has considerable potential in the remediation of heavy metals and radionuclides. Biogeochemical reactions can control the transport properties of complexed metals in the subsurface, allowing their immobilisation or subsequent dissolution and ultimate removal. However, bacterial interactions with metals are complex, and the factors influencing bacteria-matrix interactions are ill-defined. This, and a lack of information on the behaviour and transport of injected organisms in complex subsurface matrices has limited the successful employment of bio-augmentation as a remedial strategy for heavy metal and radionuclide contamination.

The research presented here focuses on laboratory and field investigations into the factors affecting the subsurface transport of augmented and indigenous organisms. The focus area for these investigations is the South Oyster Field Site located in Oyster, Virginia. This field site presently consists of two flow cells. The first flow cell (Narrow Channel focus area or NCFA) has been characterised as a relatively homogeneous, sandy, aerobic aquifer (Green et al., 2000). The second flow cell (Sub-Oxic focus area or SOFA) has been characterised as a peat to gravel heterogeneous sub-oxic aquifer (Hubbard et al., 2000). An initial field scale bacterial injection using an adhesion deficient mutant of the indigenous isolate DA001 *(Comamonas sp.)* was conducted at NC Focus Area (Mailloux et al., 2000). The bacterial plume was monitored using several novel techniques (Fuller et al., 2000a, Fuller et al., 2000b and Johnson et al., 2000). Further novel fluorescent labelling and detection methodologies for tracking injected bacteria are being developed.

Results indicated that the limited physical heterogeneity at the NCFC appears to strongly influence the transport of bacteria and a conservative tracer (Br^-). In addition, laboratory studies have been undertaken to allow parameters that appear to control the field-scale transport of bacteria to be systematically controlled. The adhesion deficient variant of DA001 and an adhesion deficient variant of an iron reducing bacterial (IRB) isolate, FER-A1E, were selected for these laboratory scale investigations. Both organisms were injected into re-packed cores containing sediments with varying physical and chemical (e.g. Fe, Al content) properties that simulate sediment physiochemical data obtained using electron microscopy techniques. The results obtained provided information on the effect of mineralogy, physical heterogeneity and the significance of chemical composition of the sediment on the 1-D transport of these bacterial strains. These investigations were performed in the field at both the aerobic and anaerobic site, allowing both NCFA and SOFA groundwater to be employed from varying depths in the two aquifers. This approach mitigated the problem of transporting actual groundwater (e.g. degassing and storage phenomenon) and more closely replicates *in-situ* conditions, in addition to providing additional information on the significance of groundwater chemistry.

This type of *in-situ* approach provides us with valuable information about bacterial-matrix interactions that can be extrapolated to field scale investigations. This ultimately improves both the understanding of bacterial subsurface transport and the effectiveness of bio-augmentation as a remedial strategy for the remediation of metals and radionuclides.

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