Geochemical zonation linked to microbial spatial distribution and microbe-mineral interactions in metal-immobilizing systems

Jonathan (Josh) Sharp (jsharp@mines.edu) - Colorado School of Mines (PI), S.M. Webb and J.R. Bargar - Stanford Synchrotron Radiation Lightsource (Co-PIs).

Recent developments in microbial community sequencing and visualization techniques enable analyses of biogeochemical zonation and microbe-metal interactions at an unprecedented detail when combined with advanced geochemical analysis and redox chemistry mapping techniques. To this end, a series of SRBRs containing solid organic substrate at different ratios have been established to query the biogeochemical zonation of microbe-mineral interactions associated with zinc immobilization. Our approach, which integrates Illumina sequencing, species-specific metallo-labeling of microbial cells and detailed synchrotron and microscopic geochemical analyses, was used to establish correlations between microbial populations and geochemical reactions.

Examination of the sulfate-reducing clades in pilot-scale systems that effectively immobilized soluble Zn revealed that the genera Desulfosporosinus and Desulfurispora coincided with high Zn-removal. In contrast, systems where the major sulfate-reducers were related to Desulfosporomusa consistently removed less Zn. Preliminary data also suggest that a similar pattern wherein phylogenetic affiliations serve as indicators for metal immobilization may exist for key cellulose degraders, such as Dysgonomonas that were negatively correlated with Zn-removal in these systems.

After approximately eight months of operation, Zn-removal was situated in the upper portions of the down-flow columns followed by a trend of downstream migration after twelve months. Canonical correspondence analysis revealed that Zn-removal was positively correlated with organic substrates containing alfalfa hay, whereas the opposite was shown for columns containing woodchips. A combined approach using scanning-electron microscopy and energy dispersive X-ray spectrometry to analyse column substrates revealed preferential associations of Zn and S in those that more effectively removed Zn over a period of one year. Further investigation employing synchrotron-based X-ray absorption spectroscopy (µXAS) is underway in order to analyze sulfur speciation and provide 2-D redox mapping of precipitates on beamline 14-3 at the Stanford Synchrotron Radiation Lightsource.

Metallo-labeling of specific microbial genera using biotinylated 16S rRNA targeted DNA-probes, followed by incubation with a streptavidin-nanogold conjugate and subsequent gold-enhancement (Gold-FISH) has been improved by targeting multiple ribosomal sites, thus significantly increasing the signal to noise ratio without severely affecting the redox chemistry of the microbe-mineral interface. We have confirmed that our improved protocol works well for Escherichia coli, Shewanella oneidensis and S. putrefaciens, the latter two are all known metal-reducers and thus highly relevant in a contaminant mobility context. Efforts are currently focused on visualization of the microbe-metal interface through Gold-FISH labeling of Shewanella cells in the presence of manganese oxide (Birnessite) and subsequent µXAS analysis.