Biogeochemistry of Uranium Under Reducing and Re-oxidizing Conditions: An Integrated Laboratory and Field Study

Brent M. Peyton[1] (PI), Rajesh K. Sani,[1] Gill G. Geesey,[2] Mary Kaufmann,[2] Katie Reardon,[2] Zbigniew Lewandowski,[2] Haluk Beyenal, [2] James E. Amonette,[3] and David Cummings

[1]Washington State University, Pullman, Washington; [2]Montana State University, Bozeman, Montana; [3]Pacific Northwest National Laboratory, Richland, Washington; Idaho National Engineering and Environmental Laboratory, Idaho Falls, Idaho

Reduction and subsequent precipitation of uranium (U) and chromium (Cr) by sulfate-reducing bacteria (SRB) and dissimilatory iron-reducing bacteria (DIRB) is well known. The use of SRB also has the potential to immobilize other toxic metals because of the large number of metals (Hg, Pb, Cd, Cu, Ni, Zn) that form stable sulfide compounds.

This project builds on our prior research to integrate laboratory and field studies focused on the NABIR FRC at Oak Ridge, TN. At the FRC, in-situ coupon experiments were conducted in a well penetrating the saturated zone of Area 3, an acidic, uranium-contaminated zone at the FRC. Hematite mineral surfaces were exposed to groundwater and indigenous flora. The bacterial community that developed over a 2-month period on hematite surfaces contained 13 Operational Taxonomic Units (OTUs) that fell primarily within the β- and γ-Proteobacteria, with over half of the recovered clones represented by a strain that exhibited 95% similarity to an *Alcaligenes* sp. and approximately 50% of the remaining clones represented by a strain that exhibited 96% similarity to a *Frateuria* sp. These strains were not represented in the more phylogenetically-diverse microbial community that colonized hematite mineral surfaces recovered from an uncontaminated site at the FRC. We developed a Ag staining method for imaging of reducing zones on mineral surfaces, including Fe oxides. To our knowledge, this is the only microscopic technique that can image Fe(II) zones on the surface of an Fe-bearing mineral with monolayer sensitivity.

In addition to field studies, laboratory experiments have been a combination of batch studies and under flowing conditions to better represent aquifer conditions for U(VI) reduction. *Desulfovibrio desulfuricans* was grown in a flat plate reactor using lactate. Uranium, in form of soluble U(VI), was continuously fed to the reactor at a concentration of 30 mg/L for eight (8) months, during which time U(VI) was completely removed from solution. Bulk water chemistry and microbial activity measurements indicate that uranium was immobilized by two simultaneous processes: (1) chemically, by reduction and precipitation with microbially generated hydrogen sulfide, and (2) and by direct reduction. The Fe(III)- and U(VI)-reducing bacterium, *Shewanella oneidensis* was grown in six fracture flow reactors (FFRs) of different geometries. The spatial and temporal distribution of substrate (tracked using a tracer dye (brilliant blue FCF)), was greatly influenced by attached growth. In all FFRs, bacterial density was observed higher at the entrance, likely due to the presence of higher electron donor concentrations. Results suggest that spatial heterogeneity of attached bacteria depends on the geometry of the fracture zone. These data will provide a baseline for developing nutrient delivery strategies for improving spatial distribution of microbial activity and U(VI) reduction capability in fracture flow systems.

Our earlier assessment of Al toxicity to SRBs was extended to include the effects of Al speciation. Thermodynamic speciation calculations and Al-27 nuclear magnetic resonance spectra provided evidence for the presence of colloidal clusters of the Al-13 tridecamer ion in the suspensions yielding the greatest decreases in SRB population.