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Insights into Stress Ecology and Evolution of Microbial Communities from Uranium-Contaminated Groundwater Revealed by Metagenomics Analyses

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Due to the uncultivated status of the majority of microorganisms in nature, little is known about their genetic properties, biochemical functions, and metabolic characteristics. Although sequence determination of the microbial community 'genome' is now possible with high throughput sequencing technology, the complexity and magnitude of most microbial communities make meaningful data acquisition and interpretation difficult. Therefore, we are sequencing groundwater microbial communities with manageable diversity and complexity (~10-400 phylotypes) at the U.S Department of Energy's Environmental Remediation Science Program (ERSP)-Field Research Center (FRC), Oak Ridge, TN. The microbial community has been sequenced from a groundwater sample (FW106) contaminated with very high levels of nitrate, uranium and other heavy metals and pH ~3.7. Consistent with trends expected in stressed ecosystems, the metagenome reveals a community of low species and strain diversity dominated by a single *Frateuria*-like γ -proteobacteria with other γ - and β -proteobacteria present at low proportions. Metabolic reconstruction reveals specific adaptations to the geochemical conditions of FW106 including genes encoding metal resistance (*czcABC*, *czcD*, *cadA*, *merA*, *arsB*), denitrification, and solvent resistance (1,2-dichloroethene, acetone, butanol). In addition to the presence of these specific genes, certain resistance genes also appear to be overrepresented in the metagenome including genes from nitrate/nitrite transport (*narK*) and metal translocation (*czcABC*, *czcD*, *cadA*), likely due to a combination of gene duplication and lateral gene transfer. A screen for positive selection shows most of these genes to be under strong negative selection, suggesting that in the short term at least, the overabundance of these transporters provide a positive fitness benefit to the cell by increasing the rate of ion transport. SNP analysis revealed a low level of polymorphism with the overwhelming majority of SNP representing unique changes within the assembled reads, suggesting that the strains in the sample are largely clonal. A model is presented for the evolution of microbial communities under high-stress conditions. To understand the metabolic

diversity of the groundwater microbial community, the microbial community (~400 phylotypes) from the background well at the FRC is also currently under sequencing.