Microbial response to spatial and temporal dynamics of uranium, iron, sulfur in ethanol-amended sediments

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We observe a significant, long-term impact in the structure of microbial communities in sediment microcosms from a uranium-contaminated aquifer that were biostimulated with ethanol, despite the amendment's rapid depletion. Sediment and groundwater samples were collected from the IFRC site adjacent to the former S-3 ponds at Oak Ridge National Laboratory. These sediments contained 713 mg kg^{-1} uranium, initially present entirely as U(VI). Ethanol (3 mM) and sulfate (3 or 9 mM) were added to the overlying water and allowed to diffuse into the sediment. U(VI) was rapidly reduced to U(IV) over a period of 40 days in the top layer of sediment in biostimulated mircocosms, concomitant with the complete consumption of added ethanol. In unamended controls, the distribution of U(VI) remained unchanged over this same time period. Following the rapid depletion of ethanol in the amended systems, both amended and unamended microcosms entered a second phase of gradual sulfate depletion coupled to U(VI) and Fe(III) reduction. Samples of the microbial community for analysis via 16S rRNA amplicon pyrosequencing were nearly four years after biostimulation. These results show the addition of ethanol led *Geobacter* sequences to predominate at the sediment-water interface, where they comprised 54-57% of all sequences compared to only 1-2% in unamended samples. Conversely, sequences related to known sulfate reducers (e.g *Desulfovibrionaceae* and *Desulfobulbaceae*) are much more abundant in this sediment layer in unamended samples (27-29%) than in biostimulated ones (4-7%). We observed little difference, however, in the community composition of sediment in the interior of the microcosms. Our results show that the strong impact biostimulation with ethanol has on microbial communities can persist long after the amendment itself has been depleted, and that this perturbation can shape microbial community dynamics for years afterwards.