SLAC SFA: Diversity and biogeography of subsurface nitrogen-cycling communities at uranium-contaminated DOE legacy sites in the upper Colorado River Basin

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The products of nitrification (NO_2^-, NO_3^-) and denitrification (NO_2^-, NO, N_2O) have been shown to impact the fate and mobility of uranium (U) at contaminated DOE legacy sites. By converting ammonia released by the decomposition of organic matter to nitrite and nitrate, nitrification effectively links the most reduced and oxidized pools of nitrogen in the environment. Under anoxic conditions, denitrification converts these nitrogen oxides to gaseous forms, which are lost from the system. Furthermore, the cycling and bioavailability of N in subsurface ecosystems is frequently coupled to redox cycles of other biogeochemical critical elements (e.g. C, S, Fe). Despite its importance, little is known regarding the diversity, abundance, and activity of N-cycling communities in such ecosystems.

We have examined ammonia-oxidizing and denitrifying microbial communities at multiple U-contaminated legacy sites in the Upper Colorado River Basin (CRB). Cores (~5-10 m deep) were collected from the alluvial floodplain at Naturita, CO, and Grand Junction, CO, by Geoprobe coring, and from Rifle, CO, Shiprock, NM, and Riverton, WY, by sonic rotary drilling. To examine the diversity and structure of the ammonia-oxidizing communities within and across these sites, a combination of sequencing and quantitative PCR of the *amoA* gene (encoding the α-subunit of ammonia monooxygenase) was applied to more than 300 sediment samples. This resulted in an extensive database of >1500 *amoA* sequences from both ammonia-oxidizing archaea (AOA) and bacteria (AOB). Interestingly, AOA *amoA* abundances ($8 \times 10^2 - 5 \times 10^8$ copies per g sediment) are consistently higher than those of AOB ($1 \times 10^2 - 5 \times 10^6$ copies per g sediment), suggesting that AOA play a prominent biogeochemical role in these sediments. Denitrifying communities are also being examined within naturally-reduced zone sediments using the functional genes *nirK* and *nirS* (encoding dissimilatory nitrite reductase) as molecular markers.

To better understand N-cycling microbial community dynamics in U-contaminated field sediments, flow-through columns were used to provide time-resolved DNA/RNA samples coupled to detailed geochemical analyses. Nitrate, nitrite, and oxygen were added to influents to study their impact on uranium oxidation. N-cycling microbial communities were characterized as a function of depth, distance from the sediment-water interface, and through time, to determine where and to what extent uranium oxidation by these oxidants occurs. Ultimately, this combination of field- and laboratory-scale approaches will provide critical insights into the interplay between microbial N-cycling and U mobility in the subsurface.