Examining the Microorganisms Assimilating Carbon from 1,4-Dioxane in Contaminated and Uncontaminated Samples

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Background/Objectives. It is well recognized that there is a critical need to develop management strategies for 1,4-dioxane due to its widespread occurrence. This chemical, a probable human carcinogen, was commonly used as a stabilizer in 1,1,1-trichloroethane formulations and is now frequently detected at sites where the chlorinated solvents are present. A major challenge in addressing 1,4-dioxane contamination concerns chemical characteristics that result in migration and persistence. Also, traditional remediation methods are largely ineffective and ex situ methods can be costly at high concentrations. Given the limitations associated with traditional remediation methods, interest has turned to bioremediation to address 1,4-dioxane contamination. Although many isolates capable of 1,4-dioxane biodegradation have been characterized, less is known about the microorganisms responsible for biodegradation in mixed community samples. The objectives of the current study were to

1) examine removal rates of 1,4-dioxane in three mixed microbial communities with different amendments,
2) identify the microorganisms responsible for the uptake of ^{13}C from 1,4-dioxane, and
3) measure and predict the functional genes present and correlate their presence to specific phylotypes.

Approach/Activities. Laboratory microcosms with media and two concentrations of 1,4-dioxane were inoculated with agricultural soil, wetland soil and sediment from a contaminated site. The impact of additional amendments on 1,4-dioxane removal rates was also examined (yeast extract and nutrients). 1,4-Dioxane concentrations were monitored over time using a triple quadrupole GC/MS system (Agilent 7010B) equipped for solid phase micro extraction. Extracted DNA was subject to stable isotope probing (SIP), involving ultracentrifugation, fractionation and 16S rRNA gene sequencing (Illumina). Additionally, both the extracted DNA and the ultracentrifugation fractions were subject to quantitative PCR targeting the functional genes associated with 1,4-dioxane biodegradation. Further, PICRUSt2 was adopted to predict the occurrence of a wider number of monooxygenases (those encoding for propane, methane, ammonia and toluene oxidation).

Results/Lessons Learned. 1,4-Dioxane removal rates were positively impacted by the presence of the additional amendments. Biodegradation rates varied between the three microbial communities. Limited removal was noted in the abiotic controls indicating biodegradation was the key removal mechanism. The SIP approach is still ongoing and should be complete within one or two months. The data that will be generated on the phylotypes responsible for carbon uptake (as determined by SIP) could be incorporated into diagnostic molecular methods for site characterization. Further, the inclusion of amendments for in situ bioremediation could reduce the time for site cleanup.