A systems biology characterization of mercury-methylating synthetic model communities

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Bioaccumulating methylmercury (MeHg) contamination is a major concern in aquatic ecosystems. Understanding the biological and geochemical factors that influence MeHg production is necessary for predicting the impacts of contamination and remediation efforts on natural systems. The *hgcAB* genes that drive MeHg production are widespread in anaerobic environments, but Hg methylation rates, kinetics, and extents have only been measured in pure cultures or *in situ* communities. It is unknown how Hg-methylation and overall cellular metabolism are affected by metabolic alterations due to syntrophic or competeitive interactions, or how those inter-species relationships affect *hgcAB* distribution and MeHg production in the field.

To address this knowledge gap we have created synthetic model communities informed by natural Hg-methylating populations in the Oak Ridge National Lab (ORNL) East Fork Poplar Creek study site, a natural stream that has been contaminated with Hg from upstream sources. Ongoing efforts at ORNL have identified active Hg methylation in EFPC stream margin sediments. We selected cultured species with >99% similarity to abundant and prevalent species in the Hg-methylating communities based on 16S, *hgcAB*, and metagenomic sequence data. The selected strains represent different anaerobic functional groups (iron and sulfate reduction, fermentation, syntrophy, methanogenesis) to simulate complete anaerobic carbon degradation. Strains were grown under steady state conditions in single culture and increasingly complex co-cultures with measurements taken of cell counts, Hg methylation rates, electron donors and acceptors, organic acids, H_2 and CO_2 concentrations, carbon balancing, and expression levels. This suite of analyses has allowed us to directly characterize the phenotypic effects of multi-species interactions on Hg-methylation and cellular metabolism in these field-informed synthetic communities.