New Insights into the Factors Defining the Composition of Methane-oxidizing Communities and the Role of Oxygen

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Project Goals: This project focuses on developing model microbial communities active in methane utilization. Two approaches are utilized: a top-down approach that employs natural communities from environmental samples, and a bottom-up approach that employs axenic cultures of model bacteria. Through manipulations of these communities, we are striving to understand the molecular mechanisms that form a basis for specific species interactions in microbial oxidation of methane. Transcriptomics and metabolomics are among the approaches that are used to identify candidate factors involved in observed co-occurrence patterns. Hypotheses validation will be approached via strain and community manipulations, such as site-specific mutagenesis or introduction of synthetic functional modules into the specific partner strains. Data integration will allow for metabolic and ecological modeling of such communities.

In the current phase of the project, we are addressing the nature of the proposed relationships among different physiological groups of microbes involved in methane utilization and the mechanisms underlying their specificity. In order to obtain mechanistic details into specific species interaction as part of microbial methane oxidation, we are now using both top-down and bottom-up community manipulation approaches. In the first, we manipulate natural communities using methane as the only carbon source and monitor complex community deconvolution toward dramatically simplified, low-species communities. In the second, we use pure cultures of bacteria, all originating from Lake Washington sediment, all with sequenced genomes, to build and manipulate simple synthetic communities. Data from the top-down approach demonstrate that, under methane pressure, the complex natural communities simplify rapidly, with a Methylococcaceae species becoming one dominant species, and a Methylophilaceae species becoming the second dominant species. However, different species persist under different conditions, one major factor being oxygen availability. At high oxygen tensions, the major players are, respectively, species of the genera Methylosarcina and Methylophilus. At low oxygen tensions, when communities experience periods of hypoxia, the major players are, respectively, species of the genera Methylobacter and Methylotenera. These data suggest that oxygen availability is a major factor determining specific partnerships in methane oxidation. Data from the bottom-up approach demonstrate that two randomly selected species of Methylococcaceae and Methylophilaceae isolated from the same environment do not readily form stable communities and that additional partners may be required, such as Flavobacteriaceae. At the same time, two-species communities were observed when both partners originated from naturally selected pairs of Methylococcaceae and Methylophilaceae. In these cases as well, oxygen appeared to play a major role in community composition and behavior. Data from both types of experiments strongly suggest that speciation within Methylococcaceae and Methylophilaceae may be responsible for niche adaptation, driven by factors such as oxygen availability. These results suggest that specific ecotypes of, respectively, Methylococcaceae and Methylophilaceae are tailored to form pairs that possess competitive advantage in specific conditions, such as the placement within the methane/oxygen counter gradient.

Publications
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