## Potential Mechanisms of Anaerobic Methane Oxidation and Challenges to Slow-Growing Microbial Communities

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## **Project Goals:**

The overarching goal to expand our understanding of the key microorganisms, metabolic strategies, and interspecies relationships involved in the formation and oxidation of methane in the environment. To study methane-cycling archaea, their syntrophic partners, and their ecophysiological properties across a range of spatial scales we will use single cell/single consortia targeted approaches combined with characterizations of field samples, geochemically-characterized laboratory microcosms, environmental bioreactors, and defined syntrophic co-cultures, and utilize novel meta'omics strategies, state-of-the-art analytical imaging methodologies and stable isotope geochemistry. Ultimately, our goal is to develop and refine predictive models of the larger scale biogeochemical processes mediated by methanemetabolizing microorganisms.

## **Abstract:**

Consortia of anaerobic methanotrophic archaea and deltaproteobacteria have been shown to oxidize methane and reduce sulfate in marine sediments, thereby limiting the rise of methane into the water column and ultimately the atmosphere. However, the mechanism of the interactions between the two groups of microorganisms remains uncertain, with recent studies (e.g., McGlynn et al. 2015, Wegener et al. 2015) pointing towards direct electron transport (DIET). Here, we use reactive transport modeling to investigate potential mechanisms of methane oxidation. To assess the potential, kinetic and thermodynamic constraints on anaerobic methane oxidation, we consider (1) the exchange of electron donors such as molecular hydrogen produced by the archaea and used by the bacteria in sulfate reduction, (2) a pathway in which the archaea directly couple methane oxidation to sulfate reduction with disulfide produced as the intermediate, which then gets disproportionated to sulfide and sulfate by the bacterial partner, and (3) direct electron transport from the archaea to bacteria through nanowires. We show that reaction kinetics, transport intensities, and energetic considerations all could decisively impact the overall rate of methane consumption. Simulation results indicate that the exchange of chemical intermediates lead to oxidation rates that fall short of measured ones. In contrast,

simulations with disulfide as intermediate and through extracellular electron transfer can achieve methane oxidation rates that match or even exceed lab measured reaction rates. To distinguish between the two mechanisms, we compare the computed spatial distribution of reaction rates to the spatial patterns within the microbial consortia as measured by <sup>15</sup>N-ammonium uptake measured with FISH-nanoSIMS. These observations show that the activities are independent of the spatial proximity of archaea and their bacterial partners in consortia. This is best reproduced in the simulations of DIET, supporting the recent evidence for such a potential mechanism.

An overall challenge in modeling microbial communities is constraining the possible range of physiology. We will address this challenge using results that define the cross-species tradeoffs in bacterial cell composition, physiological function, and metabolism, spanning five orders of magnitude in cell size (Kempes et al. 2016). Within this framework it has been shown that the largest and smallest bacteria are limited by physical space and energetic requirements (Kempes et al. 2012) and we will connect these previous efforts to define the challenges facing slow-growth microbial communities.

## References

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