

Metabolic Modeling of Synthetic Estuarine Wetlands Microbial Communities

in response to Climate Change

Dongyu Wang¹, Britt Abrahamson², Xuanyu Tao¹, Neil Q. Wofford¹, Aifen Zhou¹, David A. Stahl², Jizhong Zhou¹, Michael J. McInerney¹, Ralph S. Tanner¹, Xavier Mayali³, **Mari Winkler²**, Chongle Pan^{1*} (cpan@ou.edu)

¹University of Oklahoma, Norman, OK

²University of Washington, Seattle, WA

³Lawrence Livermore National Laboratory, Livermore, California, USA

Project Goals: Wetlands capture and release large amounts of greenhouse gases (CO₂, CH₄ and N₂O) and predicting their response to climate change induced stressors such as drought and saltwater intrusion is of prime importance. This project aims to link wetland microbial activity to ecosystem-scale processes by developing a reproducible experimental model for lacustrine and estuarine wetland ecosystems to quantify responses to controlled manipulations representing climate impacts. Hydrogel beads, controllable in size, with entrapped wetland microbes and soil plat-like, act as models for sediment aggregates. Bioreactors with real-time gas and liquid metabolite flux monitoring, integrated multi-omics analyses, and stable isotope tracing will be conceptually incorporated into mathematical models to predict how climate change stressors impact C and N fluxes across different wetland spatial and temporal scales.

Abstract:

Microbial communities in estuarine wetlands work in concert to degrade lignocellulosic biomass anaerobically to CH₄, CO₂, and H₂S. The greenhouse gas (GHG) emissions from estuarine wetland ecosystems will be severely perturbed by increased seawater intrusion caused by climate change. It remains a challenge to accurately model and predict ecosystem feedbacks to such perturbations. The altered GHG emissions from estuarine wetlands may in turn provide positive or negative feedbacks to global-scale climate change. A mechanistic understanding of metabolic networks of interaction in estuarine wetland communities is critically needed to elucidate the impact of seawater intrusion on these processes. We hypothesize that seawater intrusion of estuarine wetlands will reduce, but not eliminate, methane emissions and increase H₂S and CO₂ emissions, due to the increased availability of sulfate.

Lignocellulosic biomass in estuarine wetlands is degraded by four major functional guilds of microorganisms: cellulolytic bacteria, hydrogenotrophic methanogens, acetoclastic methanogens, and sulfate reducing bacteria. These functional guilds maintain extensive metabolic interactions with one another, including the cross-feeding of carbon from cellulolytic bacteria to the other guilds, and substrate competition between methanogens and sulfate reducers. These metabolic interactions will be perturbed by the increased sulfate availability from seawater intrusion. Sulfate respiration is energetically more favorable than methanogenesis and therefore sea level rise could promote sulfate reduction at the expense of methanogenesis and accelerate the overall carbon degradation.

Many natural wetland communities have been characterized by genotyping and meta-omics analyses. While these studies revealed the taxonomic composition and the key metabolic processes, they do not provide insights on the fate of carbon within the community due to

challenges in measuring and modeling carbon fluxes in a highly complex microbial community. There is hence an urgent need to develop a simplified mechanistic metabolic model that mimics these complex wetland communities with reduced complexity. Synthetic communities consist of defined microorganisms that represent key known metabolic processes that are representative of interactions occurring in natural communities and provide model systems of reduced complexity to study the interplays between those metabolic processes. A synthetic community also allows the integration of experimentally confirmed metabolic pathways into metabolic flux balance models that can help to better predict the carbon flux in natural ecosystems and these models can be very powerful to generate hypotheses for natural ecosystems that can be experimentally tested.

In this study, we designed a synthetic community comprised of representative members of the four major functional guilds of estuarine wetland communities that are responsible for cellulose hydrolysis, hydrogenotrophic methanogenesis, acetoclastic methanogenesis, and sulfate reduction (Figure 1). Several potential representatives were tested from each of the four functional guilds in a synthetic community, considering how they interacted *in vitro*. The most optimal consortium included the cellulolytic bacterium *Clostridium cellulolyticum*, the hydrogenotrophic methanogen *Methanospirillum Hungatei*, the acetoclastic methanogen *Methanosaeta concilii*, and the sulfate reducing bacterium *Desulfovibrio vulgaris Hildenborough*. The quad-culture was compared with a series of tri-cultures and bi-cultures of its constituents. The quad-culture was more productive in methane production than the tri-cultures and bi-cultures, demonstrating the synergy exerted by the community to conversion of cellulose to methane. The addition of sulfate to the synthetic community promoted sulfate reduction and reduced the methane production. Flux-balance models were constructed for each bacterium to simulate the carbon fluxes in the synthetic community. This provides a foundation for the development of mechanistic metabolic models of the estuarine wetland communities. We will develop and validate the metabolic models using genomics, proteomics, stable isotope probing, and gas emission data. The metabolic models will allow more accurate prediction of future GHG emissions from estuarine wetlands under climate change.

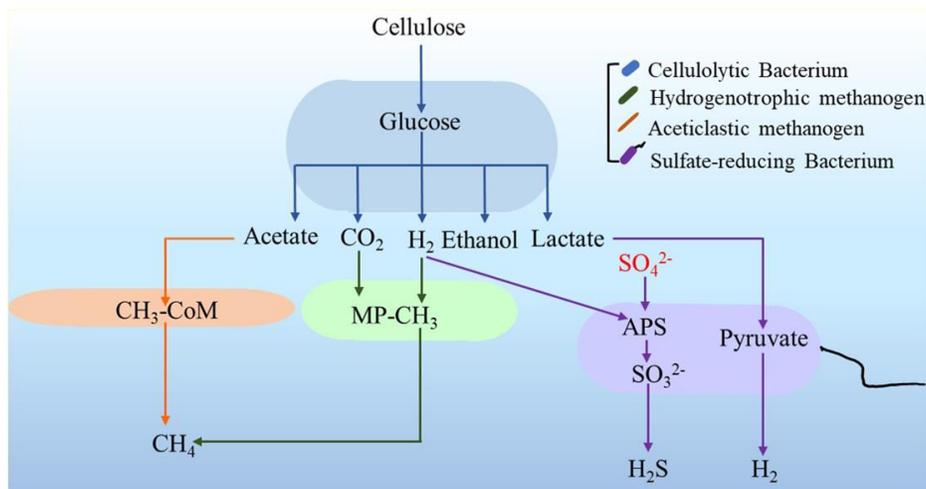


Figure1. Scheme of carbon metabolism pathway in the synthetic community

Funding statement.

This work was supported by the U.S. Department of Energy's Office of Biological and Environmental Research Genomic Science Program (Grant #DE-SC0020356); part of this work was performed under the auspices of the U.S. Department of Energy at Lawrence Livermore National Laboratory under Contract DE-AC52- 07NA27344.